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PROCEEDINGS
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*SIMULTANEOUS USE OF MOLECULAR NITROGEN AND AMMONIA BY CLOSTRIDIUM PASTEURIANUM**

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Communicated by C. A. Elvehjem, July 13, 1951

It has been shown that *Azotobacter* grown on N_2 and then supplied ammonia utilizes the ammonia to the exclusion of molecular nitrogen.[1] During investigations on the comparative biochemistry of biological nitrogen fixation, it was discovered that the nitrogen fixing anaerobe *Clostridium pasteurianum* excreted nitrogenous compounds during its active phase of growth.[2] Ammonia with extremely high N^{15} concentration was isolated from the supernatant of these cells when they were supplied N_2^{15} for a short time during the period of logarithmic growth.[3] This isolation provides direct evidence that ammonia is the key intermediate in biological nitrogen fixation

Further studies revealed that *C. pasteurianum* cells that had been fixing N_2, rapidly utilized added ammonia; excess N^{15} immediately appeared in organic compounds in the cells when the cells were supplied N^{15}-ammonia and normal N_2. These results were similar to those that had been obtained earlier with *Azotobacter*,[4] and indicated that this nitrogen-fixing *Clostridium* had enzymes for utilizing ammonia without a period of adaptation. It would be anticipated that no enzymatic adaptation would be required for utilization of a compound which is a normal intermediate in N_2 fixation. A check on the N^{15} concentration of the supernatant ammonia in these experiments showed that the atom per cent N^{15} of the supernatant decreased rapidly. This unexpected result prompted further investigations, which provide evidence that this organism once it has been fixing N_2 continues to fix N_2 even in the presence of a large excess of ammonia. This behavior helps to explain how *Clostridium* can discard newly formed ammonia while actively fixing N_2.

Experimental.—A 4-liter culture of *Clostridium pasteurianum*, strain W5, was grown anaerobically under N_2 by the methods described by Rosenblum

and Wilson.[2] When the culture was in the log phase of growth and nitrogen fixation, it was divided into 3 equal parts. At zero time, 20 ppm. of N as $N^{15}H_4^+$ containing 34 atom per cent N^{15} excess was added to each flask. N_2 was bubbled vigorously through one, H_2 through the second, and He through the third. At intervals thereafter for 2 hours, 65 ml. samples were withdrawn from each flask and pipetted into 35 ml. of N sulfuric acid. The cells then were heated rapidly to boiling to insure quick inactivation. The cells were centrifuged, washed with 30 ml. of a solution of ammonium sulfate containing 80 ppm. of N to remove or exchange with any occluded

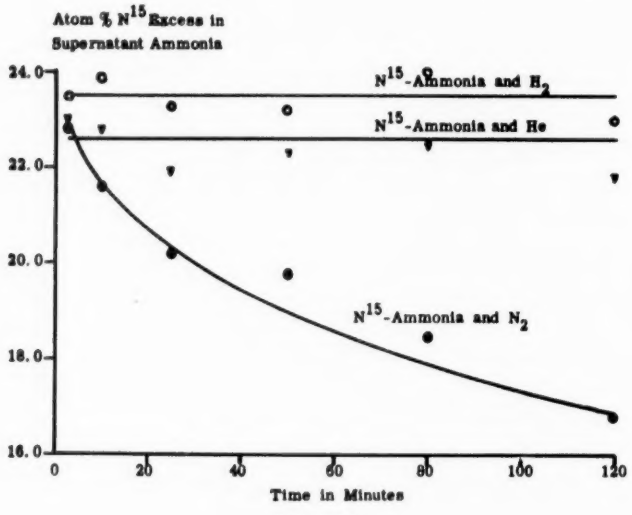

FIGURE 1

Decrease in atom per cent N^{15} of added N^{15}-ammonia by *Clostridium* also supplied N_2. Nitrogen fixing cells in the log phase of growth were given the gas indicated and 20 ppm. of N as N^{15}-ammonia.

ammonia, and then the ammonia was distilled. The supernatant medium was made alkaline, and its ammonia was distilled and recovered. The residual supernatant contained the non-ammonia supernatant nitrogen. All analyses for N^{15} were made in duplicate with a Consolidated-Nier isotope-ratio mass spectrometer, and the average of the determinations is shown in the figures.

Analyses of supernatant ammonia are plotted in Figure 1. Normal ammonia, present in the supernatant and soluble portion of the cells, diluted the ammonia added to an initial concentration of about 23 atom per cent N^{15} excess. In the presence of H_2 or He the N^{15} atom per cent

excess was constant. The He control indicated that H_2 had no specific effect on the phenomenon. In the presence of N_2, the N^{15} concentration fell from 23 to 17 atom per cent excess in 120 minutes.

Figure 2 represents the change in the ammonia-N fraction during the course of the same experiment. The supernatant ammonia-N remained constant within the error of analysis (calculation by isotope dilution indicates that about 2.5 mg. ammonia-N per liter disappeared) when either He or H_2 was bubbled through the medium containing $N^{15}H_4^+$. However, in the flask in which N_2 was bubbled through the culture, there was a marked increase in ammonia-N. This increase in ammonia-N accounted for the decrease in N^{15} concentration of the supernatant ammonia within 2%. Apparently, in spite of the addition of ammonia, nitrogen fixation

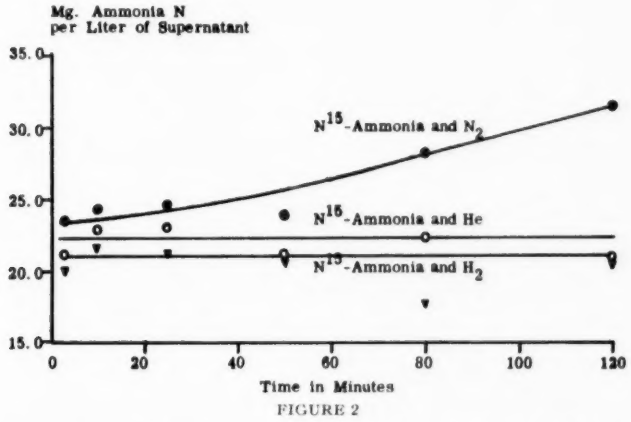

FIGURE 2

Increase in total ammonia-N in a culture of *Clostridium pasteurianum* in the presence of added ammonia and N_2.

still was taking place and was rapidly producing non-labeled ammonia. The rest of our data confirm this conclusion.

Figure 3 shows the concentration of N^{15} in the cells. The rate of assimilation of N^{15} was much more rapid in the presence of He and H_2 than in the presence of N_2. This would be expected if the added $N^{15}H_4^+$ were competing with the normal ammonia formed in the cells by nitrogen fixation in the culture supplied N_2.

Figure 4 illustrates the increase of N^{15} in the non-ammonia supernatant nitrogen. The higher content of the labeled nitrogen in this fraction in the presence of H_2 or He provides additional evidence that utilization of ammonia and fixation of N_2 are occurring concomitantly in the culture given N_2.

As a further test of the hypothesis that nitrogen fixation can take place in these cells in the presence of excess ammonia, two 1-liter cultures of *C. pasteurianum* were grown side by side. To one was added 200 ppm. of N as ammonium sulfate, more than the cells could assimilate during their growth, but no additional ammonia was added to the second. N_2 was bubbled through both flasks in series. When both cultures were in the log phase of growth, the gas was removed and replaced by 0.2 atmosphere of

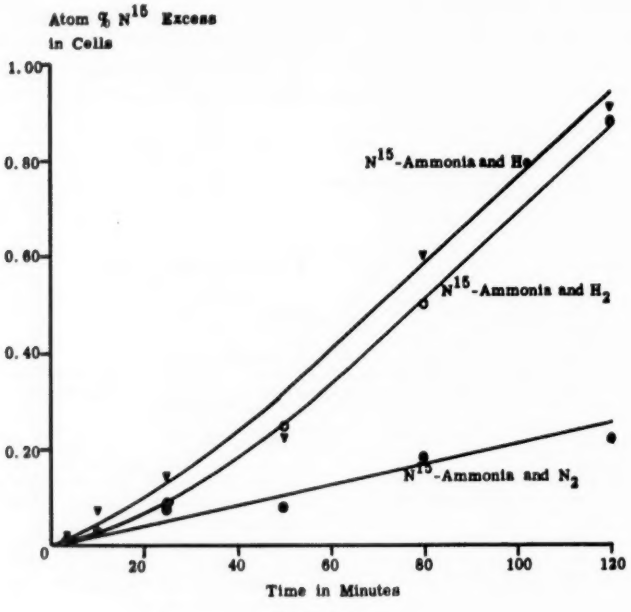

FIGURE 3

Decrease in rate of assimilation of N^{15} from N^{15}-ammonia by *Clostridium pasteurianum* supplied N_2 compared with cultures supplied He or H_2.

N^{15}-enriched N_2 (the N_2 contained 32 atom per cent N^{15} excess) and 0.8 atmosphere of He. At the same time, 20 ppm. of N as normal ammonium sulfate was added to the culture grown without added ammonia to insure an excess. The gases were bubbled vigorously through both flasks in series with a Vanton pump,[5] then samples were withdrawn from each flask at intervals and analyzed for their N^{15} concentration.

Figure 5 shows the results of this experiment. Only the cells that had been fixing N_2 continued to fix N_2 in the presence of an excess of added ammonia. Much of the nitrogen fixed was excreted rapidly, but 20 ppm. of

normal ammonia-N had been added, hence the figures for N^{15} content shown in curve *A* have been lowered by dilution. This is consistent with our earlier observation that excreted ammonia had a phenomenally high N^{15} concentration.[3]

Discussion.—There now appears to be little doubt that ammonia is a key intermediate in biological nitrogen fixation, both aerobic and anaerobic, but unlike *Azotobacter*, *Clostridium* can fix N_2 in the presence of an excess of ammonia. This fact serves to explain how *Clostridium* can excrete am-

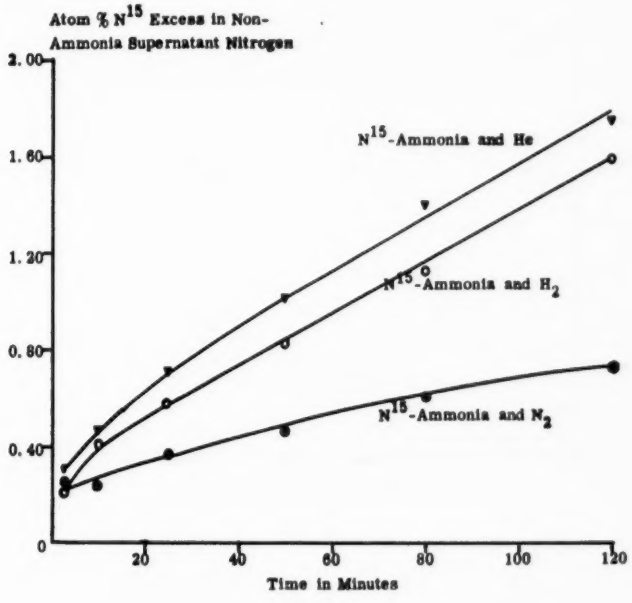

FIGURE 4

Decrease in rate of assimilation of N^{15} from N^{15}-ammonia into the non-ammonia supernatant N of a culture of *Clostridium pasteurianum* supplied N_2 as compared with cultures supplied He or H_2.

monia during active fixation of N_2. Evidently the process of N_2 fixation in *Clostridium* does not limit the rate of over-all growth, which may be limited by such factors as the quantity of ammonia acceptors produced by the organism. *Azotobacter*, however, with its vigorous aerobic respiratory system can utilize ammonia as rapidly as it is formed by the nitrogen fixing process, and there is no appreciable excretion of ammonia during active N_2 fixation.

Summary.—*Clostridium pasteurianum* cells that are fixing N_2, immedi-

ately accept ammonia as a source of nitrogen without a period of adaptation. This indicates that enzymes required for the utilization of ammonia are present in nitrogen fixing cells, and that ammonia is a normal intermediate in nitrogen fixation. When such cells are provided $N^{15}H_4^+$ and normal N_2, a substantial conversion of N_2 to ammonia occurs.

Clostridium cells that have been fixing N_2 continue to fix N_2 in the presence of added ammonia. Cells that have been growing on ammonia lack the ability to utilize N_2, and this is suggestive that nitrogen fixation itself

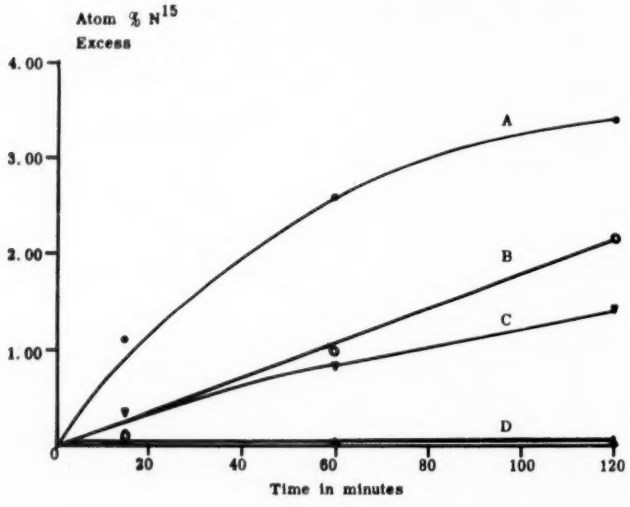

FIGURE 5

Fixation of N_2^{15} by *Clostridium* in the presence of excess added ammonia. Curve *A*—Supernatant ammonia from cells grown without added ammonia to which 20 ppm. of ammonia-N was added when N_2^{15} was supplied at zero time. Curve *B*—Cells from same experiment as curve *A*. Curve *C*—Non-ammonia supernatant nitrogen from same experiment as curve *A*. Curve *D*—Cells previously grown with 200 ppm. of N as ammonia, and N_2^{15} supplied at zero time. Curve was the same for supernatant ammonia, cells, and non-ammonia supernatant nitrogen.

is an adaptive phenomenon. These findings have been demonstrated with $N^{15}H_4^+$ and normal N_2 and with normal ammonia and N_2^{15}. The data reported here and the observation that *Clostridium* can excrete nitrogen while growing actively, help explain how it was possible to isolate ammonia with a phenomenally high N^{15} concentration in the supernatant of actively growing *C. pasteurianum*.

Acknowledgment.—The counsel of Dr. R. H. Burris and Dr. P. W. Wilson

during the course of these experiments, and the assistance of Mr. L. E. Mortenson in growing the cultures is gratefully acknowledged.

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[1] Wilson, P. W., Hull, J. F., Burris, R. H., PROC. NATL. ACAD. SCI., **29,** 289–294 (1943).

[2] Rosenblum, E. D., and Wilson, P. W., *J. Bact.*, **59,** 83–91 (1950).

[3] Zelitch, I., Rosenblum, E. D., Burris, R. H., and Wilson, P. W., *J. Biol. Chem.*, **191,** 295–298 (1951).

[4] Burris, R. H., and Wilson, P. W., *J. Bact.*, **52,** 505–512 (1946).

[5] Manufactured by Vanton Pump Corp., Empire State Building, New York, N. Y.

THE VIRUS OF VACCINIA IN CHICK EMBRYO MEMBRANE

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Communicated July 28, 1951

In any broad study of the way viruses grow within infected cells, the poxes are important as a group of closely related agents which can attack the same or related tissues. Several of them can be grown on chick-embryo membranes. In an earlier paper[1] it has been shown that the individual virus particles of fowl pox can readily be seen and recognized using the electron microscope. The present note is a record of preliminary studies made to determine how easy it is to observe the elementary particles of vaccinia in these membranes and to see if these observations give an obvious clue to the way this virus proliferates.

As in the preceding experiments with fowl pox a dilution of bacteria-free virus was placed on a dropped chorioallantoic membrane of a ten-day embryo, the shell opening was closed with scotch tape and the incubation continued. Vaccinia grows so freely on these membranes that best preparations have been obtained from embryos harvested 48 and 72 hours after inoculation. Areas of obvious pox development on the excised membranes have been chosen, fixed by immersion for not more than one hour in neutral 4% formalin-saline, dehydrated by passage through alcohols and into methacrylate monomer. In this they were embedded by polymerization in a 40°C. oven. Sections cut by the thermal expansion method[2] were mounted on formvar-covered grids, the methacrylate was removed by immersion in amyl acetate and the resulting tissue lightly shadowed with palladium.

As with fowl pox the first reaction to infection with vaccinia is a proliferation of the epithelial layer of the membrane followed by a vacuolation which has not, however, appeared as drastic as that seen with fowl pox. With vaccinia as with fowl pox, the first steps in the development of the virus elementary bodies have not yet been satisfactorily observed because of uncertainties arising from the presence in undiseased tissues of particles having approximately the dimensions of virus. When virus multiplication has proceeded sufficiently the large number of particles having the di-

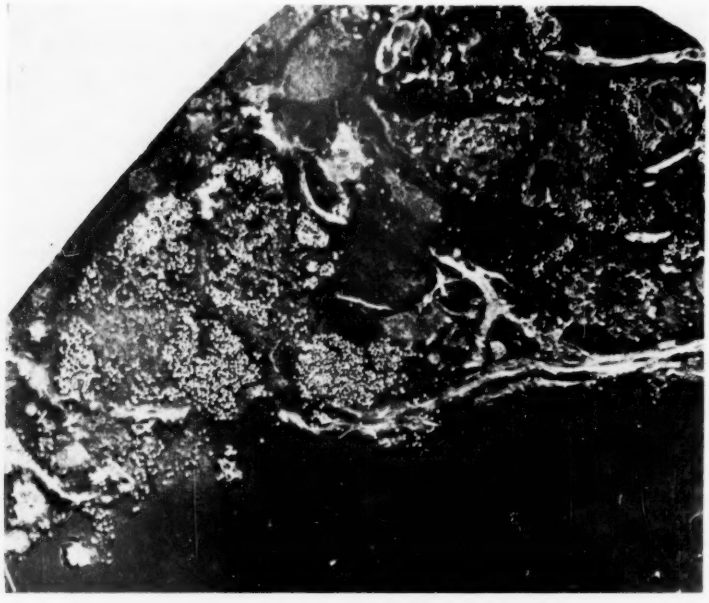

FIGURE 1

A section at low magnification showing, as minute dots, the elementary bodies of vaccinia in large clusters and small groups. In this advanced lesion normal cellular details have practically vanished. 2000 ×.

mensions of the elementary bodies in purified suspensions leaves no doubt of their viral nature.

Because they are produced in such numbers, it is easy to recognize the particles of vaccinia in infected membranes. Typical electron micrographs are shown below. The Guarnieri bodies visible under the optical microscope have long been considered as clusters of virus particles. Such clusters are readily seen at the higher magnifications attained by the electron microscope, but under these conditions we also see the virus particles dis-

tributed singly or in small groups throughout the cytoplasm of infected cells. Such virus clusters are shown at a relatively low magnification in figure 1. The association of these particles with one another can be more clearly seen in figures 2 and 3. Sometimes, as in figure 2, they are closely packed together. Even in the clusters they may be tied together by threads

FIGURE 2

A cluster of elementary bodies of vaccinia in the cytoplasm of an epithelial cell of chick-embryo membrane. Many of the virus particles are themselves sectioned. 21,000 ×.

to form a sort of coarse net replacing the original protoplasmic mesh (Fig. 3).

The present photographs do not show how the particles originate. A study of many photographs has failed to reveal forms that must be interpreted as particles undergoing division. There is a considerable variation in the size of the particles, but this may be rather a consequence of the fact

that many of them have been sectioned than an indication of a corresponding variation in their dimensions. Possibly more significant for an understanding of how this virus multiplies are the many particles which, though seemingly related to virus, are electronically less opaque and usually a little bigger (Fig. 4). They are found in cells or parts of cells where the

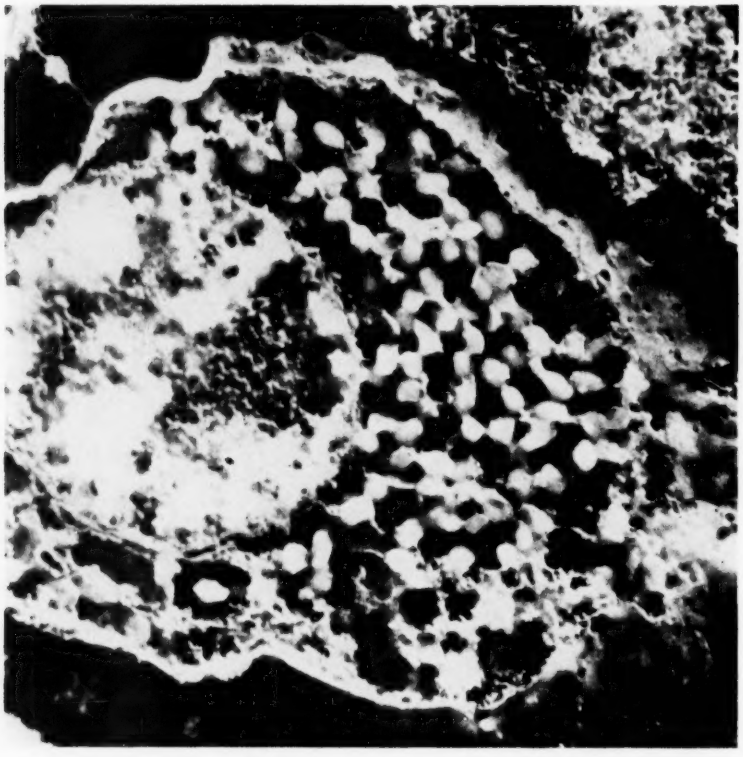

FIGURE 3

A cell in which nearly the entire cytoplasm has been replaced by a coarse net of newly formed particles of vaccinia. 17,000 ×.

original cytoplasmic net is relatively intact. They may represent earlier stages in the formation of the very dense particles we recognize as the elementary infectious units. An attempt to determine if this is indeed the case and to find out if there is any close relation between them and the few particles of similar size that can be recognized in normal epithelial cells must be a next step in the investigation of these pox viruses. Such a study

FIGURE 4

Cytoplasm of part of a cell showing small groups of (opaque) mature particles of vaccinia in the bottom half. Elsewhere are many somewhat larger less opaque objects that usually are round in outline. It will be important to determine if these are developing forms of the virus. 14,000 ×.

is now under way, using tissues infected for different lengths of time and fixed by certain reagents that might be expected to bring out cellular constituents in a preferential fashion.

[1] Morgan, C., and Wyckoff, R. W. G., *J. Immunol.*, **65**, 285 (1950).
[2] Newman, S. B., Borysko, E., and Swerdlow, M., *Science*, **110**, 66 (1949).

ANTIBIOTIC SUBSTANCES FROM BASIDIOMYCETES. VIII. PLEUROTUS MULTILUS (FR.) SACC. AND PLEUROTUS PASSECKERIANUS PILAT*

By FREDERICK KAVANAGH, ANNETTE HERVEY AND WILLIAM J. ROBBINS

DEPARTMENT OF BOTANY, COLUMBIA UNIVERSITY AND THE NEW YORK BOTANICAL GARDEN

Communicated July 18, 1951

Several species of the genus *Pleurotus* have been found in this laboratory to form substances inhibitory for *Staphylococcus aureus*. Among these were two species, *Pleurotus mutilus* (Fr.) Sacc. and *P. Passeckerianus* Pilat, obtained from the Centraalbureau voor Schimmelcultures at Baarn. An antibacterial substance formed by these fungi was isolated in crystalline form from culture liquids; it was named pleuromutilin.

P. mutilus grown on corn-steep, thiamine-peptone, or potato-dextrose agars for two days and tested by the streak-method, markedly inhibited *Staphylococcus aureus*, inhibited incompletely *Mycobacterium smegma*, and had no effect on *Escherichia coli*. Agar disks cut from colonies 10 days old formed inhibition zones 20 mm. in diameter with *S. aureus* and a small zone of incomplete inhibition with *M. smegma*. *P. Passeckerianus* produced similar zones of inhibition.

Still Cultures.—*P. mutilus* was grown at 25°C. in 2800 ml. Fernbach flasks containing beech-wood shavings and a corn-steep medium.[1] About four weeks after inoculation the mycelium covered the surface of the liquid and the activity against *S. aureus* was about 512 dilution units per ml. Reflooding the mats with fresh corn steep medium resulted in as high activity in about one week after reflooding. The reflooding was repeated at about 10-day intervals until the mat became so thick that the operation was difficult.

The flasks inoculated with *P. Passeckerianus* reached an activity of 256 dilution units per ml. in about one month.

Shake Cultures and Nitrogen Nutrition.—The fungi were also grown in 200 ml. of solution in 500-ml. Erlenmeyer flasks on a rotary shaker. In the usual corn-steep medium *P. mutilus* liquids developed an activity of 256 dilution units in about 10 days; *P. Passeckerianus* liquids were never more active than 8 dilution units per ml. Since *P. Passeckerianus* was less effective in producing antibacterial material than *P. mutilus*, the latter fungus was used in further work.

In a medium limited to corn-steep and dextrose (omitting the Dox minerals) an activity of from 2000 to 4000 dilution units per ml. was obtained in from 10 to 14 days. By adding single salts of the Dox mixture to the corn-steep-dextrose medium, sodium nitrate was found to be the interfering

substance. Nitrate not only delayed the development of antibacterial activity but also reduced the maximum activity obtained. After more than 15 days' incubation the activity of the medium containing nitrate reached one-half of that of the medium without nitrate. The effect of 9.0 g. of nitrate per liter was greater than that of 3.0 or 6.0 g.

The action of the nitrate was not direct; it appeared to be effective by modifying the metabolism of the fungus. No reduction in activity was found on incubating cultures containing nitrate to which crystalline pleuromutilin had been added. Other nitrogen sources, including ammonium sulfate, asparagine and glycine, had an effect similar to that of nitrate. Glutamic acid substituted for nitrate delayed the development of antibacterial activity but the cultures to which this substance was added eventually reached an activity equal to that of those grown with corn-steep-dextrose medium. The addition of nitrate or other nitrogen sources to the corn-steep medium had no marked effect on growth.

The mycelium grown on the shaker contained a small amount of alcohol-soluble antibacterial substance.

No evidence of a change in the capacity of the fungus to produce antibacterial substances was observed in serial transfers. Each flask of a set of flasks was inoculated with several ml. of the mycelial suspension from one flask in the previous set. The final set, representing the tenth transfer from shaker flask to shaker flask during a period of four months, was as active in producing antibacterial material as the first.

Isolation of a Crystalline Antibacterial Substance.—A white, crystalline substance (pleuromutilin) with antibacterial activity was isolated from the culture fluid. The culture fluid was extracted with 0.1 volume of chloroform at pH 5.3, the natural pH of the fluid; the chloroform was evaporated at low temperature. The brown gum remaining was dissolved in ether, the ether solution was extracted once with about 0.05 volume of 1 *N* sodium hydroxide solution to remove acidic substances, washed several times with water, and finally with dilute acid. (Extraction with alkali was not essential for the successful isolation of the active substance.) The ether was evaporated at room temperature and the brown gum allowed to crystallize. The crystalline mass was broken up by stirring with ether, filtered and washed with a small amount of ether. More crystals could be obtained from the filtrate. The brownish crystals were dissolved in a small amount of ethanol in which they were quite soluble; ether and Norit A were added and the solution filtered to remove carbon. The faintly colored solution was evaporated at room temperature to remove the alcohol which interferes with crystallization. The vitreous mass crystallized to form large, clear crystals, if allowed to stand for several days, or if seeded with a few crystals. Usually the mass was dissolved in a minimum of boiling ethanol and on the addition of ether, crystallization occurred.

The white crystals were removed, washed with ether and air-dried. The crystals were so slowly soluble that they could be washed with ether with little loss. About 50 mg. of crystalline pleuromutilin could be obtained from a liter of culture fluid.

White crystals were isolated from the culture fluid of *P. Passeckerianus* by the procedure followed for isolating pleuromutilin. The melting points of these crystals, of pleuromutilin, and a mixture of the two were the same. From this evidence and the relative antibacterial activities, we consider the substance from *P. Passeckerianus* to be pleuromutilin.

Chemical Properties of Pleuromutilin.—The crystals were soluble in alcohol, acetone, chloroform and ether; slightly soluble in water; insoluble in hexane. The melting point in open capillary tubes was 166–169°C. (cor.) without decomposition. The same sample remelted three successive times at 166°. The $[\alpha]_D^{20} \leq 0.05°$ (0.2% in 50% ethanol). The absorption spectrum showed a weak band at 290 mμ ($\epsilon = 21.7$) and end absorption below 230 mμ.

Pleuromutilin was a neutral compound free from halogen, nitrogen, sulfur, ash and without acetyl and methoxyl groups.[2] The molecular weight computed from the depression in freezing point of camphor was 310. Analyses of two lots of crystals were as shown:

	C	H	MOL. WT.
Sample 1	69.78	8.95	..
Sample 2	69.75	9.03	310
Computed for $C_{22}H_{34}O_5$	69.8	8.98	378

Crude pleuromutilin sublimed in the highest vacuum of the oil pump at about 140°C. The condensate did not crystallize and was not pure.

The first crystals were obtained from the contents of the fifth funnel of an eight-funnel counter-current distribution of crude gum between 50% (by volume) aqueous-methanol and a 3:2 mixture by volume of ethyl ether and hexane.

Quantitative acetylation with acetic anhydride in pyridine indicated between one and two acetylatable groups per molecular weight of 378. When bromine in carbon tetrachloride was added to pleuromutilin, hydrobromic acid was released. The permanganate test indicated unsaturation.

Antibacterial activity of pleuromutilin was not changed by boiling for 10 minutes in 0.2 *N* hydrochloric acid, nor in 0.5 *M* sodium bicarbonate (pH 8.8); the antibacterial activity was reduced to 6% of the original activity by boiling in 0.1 *N* sodium hydroxide. The chemical treatment and lack of decomposition on melting indicates that pleuromutilin is stable as compared to most antibiotic substances.

Antibacterial Activity of Pleuromutilin.—The antibacterial activities were measured by the methods used in this laboratory.[3] The results are

expressed as the minimum concentration in micrograms per milliliter that inhibit growth of the test bacteria for 24 hrs.

ANTIBACTERIAL ACTIVITY

Bacillus mycoides	125
Bacillus subtilis	8
Escherichia coli	500
Klebsiella pneumoniae	1
Mycobacterium smegma	32
Pseudomonas aeruginosa	>1000
Staphylococcus aureus	0.25

The activity was mainly against the Gram-positive bacteria.

Pleuromutilin at 0.5 mg./ml. was not antiluminescent when tested against a strain of luminescent bacteria.

The concentration of cells of *S. aureus* in the range from 1000 to 100,000 per ml. did not affect the end-point of the antibacterial tests.

Pleuromutilin was bacteriostatic for *S. aureus* at concentrations between 0.25 and 2 μg./ml. in beef extract medium and bactericidal at concentrations of 4 μg./ml. For concentrations of 4, 8, 16, 32 and 64 μg./ml., all of the bacteria were killed only after 3 days' exposure to pleuromutilin at 36°, although there was substantial reduction in number after 12 hrs.

Pleuromutilin was tested by the paper-disk method[4] for activity against 46 strains of bacteriophages and showed partial activity against only Streptococcus phage 6. Antibacterial activity was detected during the antiphage tests. Pleuromutilin was active against all of the strains of Staphylococcus, Streptococcus, Enterococcus, *B. subtilis*, 80% of the *E. coli* strains, one strain of *Eberthella typhii*, and 60% of the strains of *Vibrio cholera*, and was inactive against one strain of *Bacillus cereus*.

Antifungal Activity.—The antifungal activity of pleuromutilin was measured by the serial dilution method[5] in a peptone medium at pH 6. Pleuromutilin was inactive at 0.5 mg./ml. against *Aspergillus niger*, *Chaetomium globosum* (USDA 1042.4), *Gliomastix convoluta* (PQMD4c), *Memnoniella echinata* (PQMD1c), *Myrothecium verrucaria* (USDA 1334.2), *Penicillium notatum*, *Phycomyces Blakesleeanus*, *Saccharomyces cerevisiae*, *Stemphylium consortiale* (PQMD41b) and *Trichophyton mentagrophytes*.

Effect of Blood.—Incubation for 4 hrs. with 5% whole human blood in beef-extract broth did not reduce the activity of pleuromutilin.

Animal Toxicity.—Toxicity for mice was determined by injecting 0.5 ml. of solutions of pleuromutilin into a tail vein of 14–16 g. Carworth Farms CF1 male mice. The LD 50 of a single dose for mice was greater than 60 mg./kg. Pleuromutilin was given subcutaneously to mice twice daily for three days. There was no evidence of toxicity from a total of 4 mg./mouse.

Activity in Vivo.—Since pleuromutilin was not toxic for mice and could

be given in amounts sufficient to inhibit sensitive bacteria *in vitro*, infected mice[6] were treated. *Streptococcus hemolyticus* C-203 was chosen as the infecting organism since it was inhibited by 0.5 μg./ml. in the presence of rabbit blood. The mice were infected by a dose of one-half million cells given intraperitoneally. The treatment with pleuromutilin was started immediately after infecting the mice. The solution of pleuromutilin was given subcutaneously either as a single dose or as three doses per day for as long as the mice survived. When given as a single dose of pleuromutilin at 50 mg./kg., 63% of the mice were alive 7 days after infecting while 43% survived when given 100 mg./kg. All of the penicillin treated and 20% of the untreated controls were alive at 7 days. All of the mice died within 7 days when they were treated three times daily with either 25 mg./kg. or 50 mg./kg. doses of pleuromutilin.

Pleuromutilin at a daily dose of 400 mg./kg. did not retard the growth of sarcoma 180 in mice.[7] Pleuromutilin was inactive against *Plasmodium gallinaceum* at 40 mg./kg. when administered orally twice daily for four days.[8]

Repeated contact with solutions or solid pleuromutilin caused mild urticaria in susceptible individuals.

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[1] The usual corn-steep medium contained, per liter, 1.5 g. KH_2PO_4, 0.5 g. KCl, 0.5 g. $MgSO_4\ 7H_2O$, 3 g. $NaNO_3$, 40 g. dextrose, and 5 g. Staley special nutrient 114 (corn steep).

[2] The analyses were made by Mr. J. F. Alicino and by The Huffman Analytical Laboratories.

[3] Kavanagh, F., *Bull. Torrey Bot. Club*, **74**, 303–320 (1947).

[4] Asheshov, I. N., Strelitz, F., and Hall, E., *Brit. Jour. Exptl. Path.*, **30**, 175–185 (1949).

[5] Anchel, M., Hervey, A., Kavanagh, F., Polatnick, J., and Robbins, W. J., these PROCEEDINGS, **34**, 498–502 (1948).

[6] Through the courtesy of Dr. W. C. Robbins, of Cornell Medical School, infected mice were treated with pleuromutilin.

[7] Tests made through the courtesy of Dr. Chester Stock of Sloan-Kettering Institute for Cancer Research.

[8] Tests made through the courtesy of Dr. Joseph Greenberg of the National Institutes of Health.

IMMUNOLOGIC ADSORBENTS. I. ISOLATION OF ANTIBODY BY MEANS OF A CELLULOSE-PROTEIN ANTIGEN*

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For the past several years we have been interested in devising methods that would afford practical procedures for the isolation and purification of antibodies from immune serums and in particular the isolation of non-precipitating antibodies from animal serums and allergic antibodies (reagins) from human serums.[1] The general approach has been to attempt to produce an insoluble protein antigen which would combine specifically with antibody to give a complex that could be dissociated into soluble antibody and insoluble antigen, which could then be separated by centrifugation. Attempts were made to confer insolubility on protein antigens such as ovalbumin and crystalline bovine serum albumin without destroying their antigenicity. Products were obtained by use of the usual denaturing agents, tannic acid, and bifunctional coupling reagents (e.g., tetrazotized benzidine), and also by coupling to an insoluble substrate such as phenolic resins or insoluble proteins (such as fibrin). However, in every case the resulting product either had lost too much native antigenicity (i.e., adsorbed serum always contained antibody which reacted with native antigen), was too soluble, or had too much power of adsorption for non-specific protein.

A most promising antigen has now been prepared by coupling protein to a powdered cellulose (Solka-floc). The general method of synthesizing the cellulose-protein compound consists in conversion of the cellulose to a *p*-nitrobenzyl derivative, reduction of the nitro group, and subsequent diazotization and coupling to protein in the usual manner.

p-Nitrobenzylcellulose.—Powdered cellulose (Solka-floc) was thoroughly washed with dilute acid, dilute alkali, and water, and then dried. Four grams of this material was then mixed with 12 g. of *p*-nitrobenzylchloride and 30 ml. of 40% sodium hydroxide and stirred vigorously at 95°C. (The reacting mixture was cooled during the first part of the exothermic reaction.) After 4 hrs. the mixture was poured into a large excess of cold water and filtered, and the residue was washed with water, with ethanol, and finally with acetone in a Soxhlet extractor. The product contained about 1.5 benzyl groups per glucose unit, and the yield, based on the cellulose used, was close to 100%.

p-Aminobenzylcellulose.—Five grams of the nitro compound was suspended in 50 ml. of ethanol and heated to near boiling. The mixture was then stirred vigorously and 5 g. of sodium hydrosulfite ($Na_2S_2O_4 \cdot 2H_2O$) dis-

solved in water was slowly added. After continued heating of the mixture for about 30 minutes the light yellow product was filtered off and washed with cold water.

Diazotization.—Five grams of the *p*-aminobenzylcellulose was suspended in 10 ml. of 2 *N* HCl, then mixed with 20 ml. of water and chilled in an ice bath. With constant stirring, 0.5 *M* $NaNO_2$ solution was slowly added until a test with KI-starch paper remained positive for 15 minutes after addition of the last portion of nitrite. Stirring was then continued for another 15 minutes and the material was filtered and washed with weakly acid ice water.

Coupling to Protein.—The precipitate of diazonium salt was added to ice-cooled 2% solution of protein in borate buffer pH 8.75. The protein used was crystalline bovine albumin (Armour). The amount of dry protein used for the coupling reaction was generally one-fourth to one-fifth of the dry weight of *p*-aminobenzylcellulose. The coupling mixture was stirred at low temperature for 2 hrs., the pH was adjusted to 7.3, and the mixture was stored at 4°C. for at least 36 hrs. The product was then filtered and washed with large amounts of buffer. The amount of coupled albumin in the preparation used in the present study was about 1.5% of the total weight of the adsorbent.

Covering Unreached Diazonium Groups.—Diazobenzylcellulose seems to be a very stable compound; even after 50 hrs. unreacted diazonium groups are detectable in the coupling mixture. In order to prevent their interaction with serum proteins during adsorption experiments the preparation was allowed to react with β-naphthol, forming a dark red azo dye; proteinazobenzylcellulose was stirred for 30 minutes in 10 volumes of a saturated ice-cooled solution of β-naphthol in borate buffer of pH 8.75, followed by renewed washing on filter with buffer and water.

Since the color of the protein-containing product was only faintly brownish, the reddish tint of the naphthol dye was a qualitative criterion for the completeness of the reaction with the protein. It was assumed that steric hindrance made it impossible for the albumin molecules to couple with all available diazo groups, these being, on the other hand, easily reached by the much smaller naphthol molecule.

The cellulose-albumin complex was found to be an efficient specific adsorbent. Non-specific (non antibody) serum proteins could easily be washed from the adsorbent with 1.0% saline solution. We also found that the chromatographic column had many advantages over the suspension technique, especially for removing all antibodies, including non-precipitating or weakly combining types.

A disconcerting reaction with some preparations of insoluble antigens was the lack of complete removal of all antibody from a sample of serum. This might have resulted from the involvement of certain native haptenic

groups in the coupling reaction or their destruction in some other manner. The cellulose-antigen complexes made with careful control of pH and other coupling conditions, do remove all antibody. Drying the product caused some loss of specific activity.

The antiserum was a pool obtained from rabbits after three to six months immunization with bovine serum albumin (from the batch used for the tests). It contained about 3.39 mg. of precipitating antibody per ml., as determined by the method of Lanni and Campbell.[2]

A 5.0-ml. sample of this serum (containing 17.0 mg. of precipitating antibody) was diluted with 5.0 ml. of 1.0% saline and allowed to pass slowly through a column of about 1.0 g. (wet weight, 0.46 g. dry weight) of the cellulose albumin adsorbent. The packed column was approximately 20 × 8 mm. in size. About 2 hrs. at room temperature was required for the 10 ml. of diluted serum to pass through by gravity only. At the end of this time a small sample of filtrate was removed for antibody determination and the remaining solution was passed through a second similar column. The filtrate from the second column contained no detectable antibody. Analysis of the first filtrate gave a value of about 7.20 mg. total precipitating antibody not adsorbed. This amounts to about 40% not absorbed by the gram of wet adsorbent. If no native antigenic groups had been destroyed in preparation of the adsorbent one would expect that the remaining 40% of antibody could be removed by passage through another column of similar dimensions, which was in fact accomplished.

The acid dissociation method reported by Campbell and Lanni[3] was used to elute the antibody from the adsorbent. The adsorbent was removed from the two chromatographic tubes, suspended in 1.0% saline, and washed and centrifuged three or four times, until the washings were biuret-negative. The pH was then adjusted to approximately 3.2 with 0.1 *N* HCl and the suspension was stirred gently for 60 to 90 minutes at room temperature. The insoluble residue was then resuspended in 1.0% saline and the pH adjusted to 8.2 with borate buffer and tested for precipitating antibody.

Earlier experiments in which denatured albumin was used indicated that elution of antibody which had reacted with remaining antigenic groups was fairly simple. It has been found that ease of elution decreases with increasing length of time that the antibody is allowed to remain on the column. It is important therefore to remove the antibody as soon as possible. When columns are stored in the refrigerator for more than 2 days little or no antibody can be eluted. The following typical elution experiment was carried out with the materials described above. Two samples of 5.0 ml. each of antiserum (3.39 mg. precipitable antibody per ml.) diluted with 5.0 ml. of 1.0% saline were passed through separate columns each containing about 2.0 g. (wet weight) of adsorbent. One column was washed and the antibody was eluted immediately, and the other column

was stored at 4°C. for 12 hrs. before the antibody was eluted. No precipitating antibody could be detected in either filtrate and hence it was assumed that all 17.0 mg. of antibody had been adsorbed in each case. A total of 14.64 mg. of antibody was recovered from the column eluted at once but only 9.50 mg. from the column stored for 12 hrs. at 4°C.

Purity (precipitable antibody/total protein) of recovered soluble material obtained by specific adsorption and elution is high. In the above experiment both columns were eluted at pH 3.2 at room temperature, and the eluate was adjusted to a final volume of 25 ml. The eluate from the first column contained 0.67 mg. of protein per ml., of which 0.59 mg./ml. was precipitable with antigen. The second column gave 0.42 mg. of protein per ml., with 0.38 mg. of precipitable antibody The apparent purity was therefore slightly less than 90% in both these instances, and similar results were obtained with other preparations. It is likely that the eluted protein contains a still larger fraction of antibody, since the antigen-antibody ratio necessary for complete precipitation of antibody may not have been achieved in the precipitation tests.

The question regarding the efficiency of the regenerated (acid-treated) immune adsorbent has not been thoroughly studied, but preliminary tests have indicated that the adsorbing power after one antibody elution is not greatly different from that of the original material. One would expect, however, that some loss would be induced by denaturation of the albumin as well as by lack of complete removal of antibody.

Summary.—Methods are described for the preparation and use of an insoluble antigenic adsorbent obtained by coupling protein to diazotized *p*-aminobenzylcellulose. The data which are presented indicate the practical usefulness of such a method for the isolation and purification of antibody and suggest that such chromatographic techniques might be applied to the separation of antibodies with different reaction capacities.

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[1] Campbell, Dan H., and Lerman, Leonard, *Federation Proceedings* **8**, 402 (1949).

[2] Lanni, Frank, and Campbell, Dan H., *Stanford Medical Bull.*, **6**, 97 (1948).

[3] Campbell, Dan H., and Lanni, Frank, *Amino Acids and Proteins*, Chapt. XI, Thomas, Springfield, Ill., 1951.

THE POTENTIAL OF AN INFINITE ROD-LIKE MOLECULE AND THE DISTRIBUTION OF THE COUNTER IONS

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The Debye-Hückel theory provides an adequate description of the behavior of simple electrolytes at low concentrations. It contains, however, two inherent limitations: spherical symmetry, and the condition that the electrostatic potential energy of a test ion be small compared to kT. Several cases involving non-spherical geometry have been treated; Verwey and Overbeek[1] summarize the pertinent literature to 1948. In the case of polyelectrolytes[2] (especially in the absence of added simple electrolyte), we find high concentrations of charge in small volumes; in the vicinity of such a multiple charge, the potential energy of a counter ion is necessarily many times kT. Furthermore the geometry is in general not spherically symmetric; it has been concluded on empirical grounds[2] that a polyelectrolyte becomes an extended rod-like structure at low concentrations, due to intramolecular electrostatic repulsion. We present herewith the mathematical solution of the problem of the potential of an *isolated* rod-like molecule in the presence of an electrically equivalent number of counter ions; the distribution and density of the latter may then be derived from the potential. The derivation depends on explicit use of the Boltzmann distribution function, and assumes linear superposition of fields in a case where the Poisson equation is non-linear. The justification of these fundamental assumptions is based on a comparison with experiment,[3] details of which will be presented later.

Let us consider a rigid rod-like molecule of radius a stretched along the axis of a cylinder of radius R. Assume that the length h of the cylinder is large compared to R, so that end effects may be neglected. Suppose the rod carries ν ionogenic sites, so that the linear density of sites is ν/h per cm.; assume further that these sites are uniformly distributed along the rod. Assume the volume in the annulus to be filled with a continuous medium of dielectric constant D, containing at time zero a solute which will react with the ionogenic sites to produce ions. Let the concentration of solute be n molecules per cm.^{3}, where n is chosen so that

$$\nu = \pi n(R^2 - a^2)h. \tag{1}$$

If we allow the reaction to proceed to completion, the rod will then carry a charge $\nu\epsilon_1/h$ per cm. and the annulus will contain a charge $\pi n(R^2 - a^2)h\epsilon_2$, where $\epsilon_1 = -\epsilon_2$. Here the absolute value of ϵ_1 and ϵ_2 will be taken as the unit charge, 4.80×10^{-10} e. s. u. The n counter ions so produced will ob-

viously no longer be uniformly distributed as were the neutral parent molecules.

As a practical example, the rod might be an extended molecule containing ν tertiary nitrogen atoms per length h of chain, and the simple molecules in solution be methyl bromide; reaction will charge the chain positively and produce a cloud of negative counter ions whose local concentration becomes a function of the distance from the rod. If the rod plus counter ions is considered as a single molecule of polyelectrolyte, the volume available per macromolecule is $\pi R^2 h$. Concentrations of polyelectrolytes are frequently expressed as monomoles per liter, i.e., c is the stoichiometric concentration in moles of monomer per liter. If we assume one ionogenic site per monomer unit, $\pi R^2 h/\nu$ equals $1000/Nc$, where N is Avogadro's number. Consequently we see that R in our model is a measure of average concentration:

$$c = 10^3\nu/\pi N R^2 h \tag{2}$$

if we eventually consider a system of many rod molecules, to each of which is assigned an average volume $\pi R^2 h$.

If $\rho(r)$ is the density of counter ions, the potential $\psi(r)$ of a test ion is given by the Poisson equation

$$\Delta\psi = -4\pi\rho/D$$

where Δ is the Laplace operator in cylindrical coordinates; here, of course, it simply reduces to a function of r, where $a \leqslant r \leqslant R$. If we assume that the Boltzmann factor gives the probability of finding an ion at a distance r, then

$$\Delta\psi = -(4\pi n\epsilon_2/D)\exp(-\epsilon_2\psi/kT). \tag{3}$$

We are indebted to Professor M. Shifer of the Hebrew University of Jerusalem for suggesting the following transformation. Let

$$v = -(\epsilon_2\psi/kT) + 2\ln r \tag{4}$$

and

$$u = \ln r. \tag{5}$$

Substituting (4) and (5) in (3) and multiplying by $2dv/du$, we obtain

$$\frac{d}{du}\left(\frac{dv}{du}\right)^2 = 2\kappa^2\left(\frac{de^v}{du}\right)$$

where

$$\kappa^2 = 4\pi n\epsilon^2/DkT \tag{6}$$

The first integration can now be performed by inspection, giving

$$(dv/du)^2 = 2\kappa^2 e^v + 4C \tag{7}$$

where C is the integration constant and the factor of 4 has been inserted for later convenience. Equation (7) can also be integrated in closed form; on resubstitution of (4) and (5), the final result is

$$\psi = (kT/\epsilon_2) \ln [(\kappa^2 r^2/2C) \sinh^2 \sqrt{C}\xi] \tag{8}$$

where

$$\xi = \ln Ar$$

and A is the second constant of integration. If we let

$$|C| = \beta^2; \quad \sqrt{C} = \pm\beta, \pm j\beta \tag{9}$$

we obtain the following explicit real solutions of (7)

$$\psi_1(r, \beta) = (kT/\epsilon_2) \ln [(\kappa^2 r^2/2\beta^2) \sinh^2 \beta\xi] \tag{10}$$

$$\psi_2(r, 0) = (kT/\epsilon_2) \ln (\kappa^2 r^2 \xi^2/2) \tag{11}$$

$$\psi_3(r, \beta) = (kT/\epsilon_2) \ln [(\kappa^2 r^2/2\beta^2) \sin^2 \beta\xi] \tag{12}$$

according to whether $C > 0$, $C = 0$ or $C < 0$, respectively. As we shall see later, ψ_1 corresponds to low charge densities on the rod and ψ_2 to high charge densities. There remains the task of evaluating the constants A and β from the boundary conditions.

Suppose we consider an assembly[4] of parallel rod molecules; by symmetry, it is necessary that the potential have a minimum on an equipotential surface surrounding each rod. On this surface, the field must vanish. In order eventually to correlate our results with real systems, we shall require as the first boundary condition for our isolated model that

$$R(d\psi/dr)_{r=R} = 0 \tag{13}$$

and as the second that

$$a(d\psi/dr)_{r=a} = -2\nu\epsilon_1/hD \tag{14}$$

As stated, the second boundary condition implies that the gegen ions are point charges; assignment of a finite radius to them is a refinement which can be made later. Condition (14) is, of course, also the general condition for electrical neutrality of the system, because the following equation must be satisfied:

$$\nu\epsilon_1/h + 2\pi n\epsilon_2 \int_a^R e^{-(\epsilon_2\psi/kT)} r\, dr = 0 \tag{15}$$

Differentiation of (10) gives

$$r(d\psi/dr) = (2kT/\epsilon_2)(1 + \beta \coth \beta\xi) \tag{16}$$

and substitution in (13) and (14) gives

$$\coth \beta\xi_R = -1/\beta \tag{17}$$

and

$$\coth \beta\xi_a = (\lambda - 1)/\beta \tag{18}$$

where

$$\lambda = -\nu\epsilon_1\epsilon_2/hDkT = \nu\epsilon^2/hDkT \tag{19}$$

is a quantity to which we shall refer as the charge parameter. By combining (1), (6) and (19), we obtain

$$\kappa^2R^2/2 = 2\lambda/(1 - a^2/R^2) \tag{20}$$

The factor $(1 - a^2/R^2)$ must be retained in convergence tests, but becomes negligible at low concentrations, where we may set

$$\kappa^2R^2 = 4\lambda \tag{21}$$

It will be noted that the charge parameter and concentration are thus directly related, as indeed they obviously must be.

Boundary condition (17) and its analogs for ψ_2 and ψ_3 immediately give the potential at the boundary $r = R$:

$$\psi_1(R, \beta) = (kT/\epsilon_2) \ln [\kappa^2R^2/2(1 - \beta^2)] \tag{22}$$

$$\psi_2(R, 0) = (kT/\epsilon_2) \ln (\kappa^2R^2/2) \tag{23}$$

$$\psi_3(R, \beta) = (kT/\epsilon_2) \ln [\kappa^2R^2/2(1 + \beta^2)] \tag{24}$$

Similarly, we obtain from (18) and its analogs the potential at the rod where $r = a$:

$$\psi_1(a, \beta) = (kT/\epsilon_2) \ln \{\kappa^2a^2/2[(1 - \lambda^2) - \beta^2]\} \tag{25}$$

$$\psi_2(a, 0) = (kT/\epsilon_2) \ln [\kappa^2a^2(1 - s)^2/2] \tag{26}$$

$$\psi_3(a, \beta) = (kT/\epsilon_2) \ln \{\kappa^2a^2/2[(1 - \lambda)^2 + \beta^2]\} \tag{27}$$

In (26), after using (13) to eliminate A_2 for this case, we have introduced the abbreviation

$$s = \ln a/R \tag{28}$$

It is a measure of concentration; according to (2),

$$2s = \ln c + \ln (\pi Nha^2/10^3\nu) \tag{29}$$

We note that $s < 0$ by definition; since the potential as given in (10) and (12) is independent of the sign of β, we shall choose $\beta < 0$, so that the (frequently appearing) quantity βs will automatically be positive.

In order to eliminate A, we write (17) in exponential form and solve for $(AR)^{2\beta}$. The result is

$$\ln A = -(1/\beta) \tanh^{-1} \beta - \ln R \tag{30}$$

$$= \left(\frac{1}{2\beta}\right) \ln [(1 - \beta)/(1 + \beta)] - \ln R \tag{31}$$

Substitution in (10) and the analogous expression in (12) gives

$$\psi_1(r, \beta) = (kT/\epsilon_2) \ln \{(\kappa^2 r^2/2\beta^2) \sinh^2 [-\tanh^{-1} \beta + \beta \ln (r/R)]\} \quad (32)$$

$$\psi_3(r, \beta) = (kT/\epsilon_2) \ln \{(\kappa^2 r^2/2\beta^2) \sin^2 [-\tan^{-1} \beta + \beta \ln (r/R)]\} \quad (33)$$

We now have the potential expressed as a function of β, which is in turn a function of λ and s. Substituting (31) in (18), expanding the form coth $(x + y)$ and simplifying the result, we obtain

$$\lambda = (1 - \beta^2)/(1 - \beta \coth \beta s) \quad (34)$$

and in an analogous fashion, for the range of ψ_3,

$$\lambda = (1 + \beta^2)/(1 - \beta \cot \beta s) \quad (35)$$

Inspection of (34) shows that $\beta = -1$ corresponds to $\lambda = 0$, i.e., the limit of vanishingly small charge density. (By the convention already mentioned, we disregard positive values of β). If we allow β to approach zero ($s \neq 0$, of course) in (34) or (35), we find

$$\lim_{\beta \to 0} \lambda = s/(s - 1) = \Lambda \quad (36)$$

where

$$\lim_{s \to -\infty} \Lambda = 1. \quad (37)$$

For low charge densities on the rod, i.e., for $0 \leqslant \lambda < \Lambda$, the potential is described by $\psi_1(r, \beta)$; for high charge densities, i.e., $\Lambda < \lambda < \infty$, the potential is given by $\psi_3(r, \beta)$; for $\lambda = \Lambda$, where $\ln A_2 = -1 - \ln R$,

$$\psi_2(r, 0) = (kT/\epsilon_2) \ln \{(\kappa^2 r^2/2)[1 - \ln (r/R)]^2\} \quad (38)$$

Ideally, we should now solve (34) and (35) to find $\beta = \beta(\lambda, s)$ and then substitute the results in (32) and (33) in order to obtain $\psi = \psi(r, \lambda, s)$. The transcendentals cannot, however, be solved in closed form, and it is necessary to use β as an intermediate parameter. We shall first consider a specific example of $\beta(\lambda, s)$, and then proceed to a general discussion.

According to (8) and (36), $\sqrt{C}$ passes from real to imaginary values when the charge parameter goes through Λ. In order to show the complete course of the integration constant, we have plotted $(-\beta)$ and $(-j\beta)$ against λ in figure 1, for the round value $s = -3$. For $\lambda = 0$, β starts at -1 and decreases (in absolute value) linearly at first, and ultimately approaches zero parabolically as λ approaches Λ. At $\lambda = \Lambda$, $\beta = 0$; beyond this point, $\sqrt{C}$ becomes imaginary, so that $j\beta$ now increases parabolically at first. As λ further increases, β increases, eventually becoming asymptotic to a value β_∞ which corresponds to the root of

$$\beta_\infty \cot (\beta_\infty s) = 1 \quad (39)$$

where the denominator of (35) vanishes; this is the limit for a hypothetical infinite charge on the rod. As concentration is decreased, Λ moves upwards toward unity, and the linear range of β for $\lambda < \Lambda$ increases correspondingly. For $\lambda > \Lambda$, the imaginary arc of the curve moves in toward the λ-axis; we shall see later that $\beta_\infty \sim \pi/s$ for large values of s (low concentrations). In the limit of $|s| \gg 1$, the map of β becomes simply two straight lines, one running from $(-1, 0)$ to $(0, 1)$ and the other the (imaginary side of the) λ-axis.

Some useful limiting forms of (34) and (35) will next be derived. Rearrangement of (34) gives the mixed quadratic-transcendental

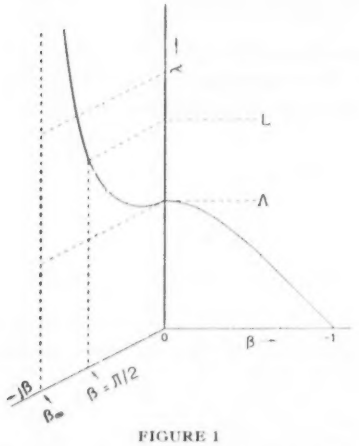

FIGURE 1

Dependence of integration constant on charge parameter.

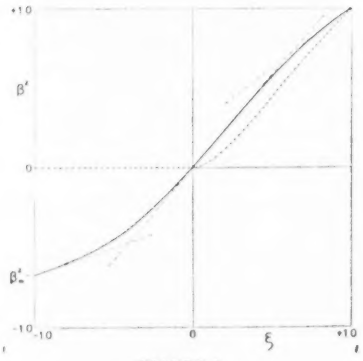

FIGURE 2

Dependence of β^2 on ζ.

$$\beta^2 - \lambda\beta \coth (\beta s) - (1 - \lambda) = 0 \tag{40}$$

If we solve (40) as a quadratic, substitute the identity

$$\coth \beta s = (1 + e^{-2\beta s})/(1 - e^{-2\beta s}) \tag{41}$$

and expand the radical in inverse powers of $(2 - \lambda)^2$, the expansion for small (i.e., $\lambda < \Lambda$) values results:

$$-\beta = (1 - \lambda)\,[1 - \lambda x/(2 - \lambda) + \lambda^2 x^2/(2 - \lambda)^3 + O(x^3)] \tag{42}$$

where

$$x = 2e^{-2\beta s}/(1 - e^{-2\beta s}) = -1 + \coth \beta s \tag{43}$$

Equations (42) and (43) permit a convenient convergent cycle of successive

approximations, starting with $-\beta \approx (1 - \lambda)$ in (43) as 0th approximation.

When λ is near Λ, the auxiliary variable

$$\zeta = (\Lambda - \lambda)/(\Lambda + \lambda) \tag{44}$$

is useful. By a limiting process to determine $dC/d\lambda$ near $(C = 0, \zeta = 0)$, we find by series expansion of (34) or (35)

$$\beta^2 \approx 6\zeta/(3 - \Lambda s) \tag{45}$$

For λ larger than Λ by amounts which make (45) unreliable, we expand around $\beta s = \pi/2$ where the denominator of (35) reduces to unity. If we set

$$L = 1 + \pi^2/4s^2 \tag{46}$$

and

$$y = \beta - \pi/2s \tag{47}$$

and approximate $-\cot \beta s = \tan ys$ in (35) by ys, the following approximation is obtained:

$$y = \{|\pi q| - [\pi^2 q^2 - 4(L - \lambda)(1 - \lambda s)]^{1/2}\}/2(1 - \lambda s) \tag{48}$$

where $q = (2 - \lambda s)/2s$. Finally, for very large values of the charge parameter, a limiting process applied to the derivative of (35) with respect to $(1/\lambda)$ gives as the leading terms

$$\beta \approx \beta_\infty + B/\lambda \tag{49}$$

where β_∞ is the asymptotic value approached by β for large values of λ and

$$B = \beta_\infty(1 + \beta_\infty^2)/[s(1 + \beta_\infty^2) - 1] \tag{50}$$

An interesting symmetry appears on comparing (42) and (49): for small values of λ, β is linear in λ; while for very large values, it becomes linear in the reciprocal of λ. The variable ζ of (44) shows the same behavior. When s is large, a first approximation to the solution of (39) which determines β_∞ is given by

$$\beta_\infty \approx (\pi/s)(1 + 1/s) \tag{51}$$

The variable ζ defined in (44) permits a very compact representation of the parameter β, because it transforms the range $0 \leqslant \lambda \leqslant \infty$ to $1 \geqslant \zeta \geqslant -1$. The solid curve of figure 2 is $\beta^2(\zeta)$ for $s = -3$. Three points and the corresponding slopes are contained in the formulas developed above: at $\zeta = 1$, $\beta^2 = 1$ and $d\beta^2/d\zeta = \Lambda$; at $\zeta = 0$, $\beta^2 = 0$ and $d\beta^2/d\zeta = 6/(3 - \Lambda s)$; and at $\zeta = -1$, $\beta^2 = \beta_\infty^2$ and $d\beta^2/d\zeta = \beta_\infty B/\Lambda$. The limit for $|s| \gg 1$ is shown as the dotted curve in the range $0 < \zeta \leqslant 1$; for $-1 \leqslant \zeta < 0$, the curve coincides with the ζ-axis, corresponding to the inverse relationship (51), where $\beta_\infty \sim \pi/s \ll 1$.

We next investigate the potential at the surface of the rod, which is given by (25), (26) and (27). So far, no approximations have been introduced into these equations, beyond those inherent in the model: they are the direct consequences of the Poisson equation and the boundary conditions (17) and (18). By substituting (20), we obtain

$$\psi_1(a, \beta) = (kT/\epsilon_2) \ln \{2\lambda e^{2s}/[(1 - \lambda)^2 - \beta^2](1 - e^{2s})\} \quad (52)$$

$$\psi_2(a, 0) = (kT/\epsilon_2) \ln [2e^{2s}s(s - 1)/(1 - e^{2s})] \quad (53)$$

$$\psi_3(a, \beta) = (kT/\epsilon_2) \ln \{2\lambda e^{2s}/[(1 - \lambda)^2 + \beta^2](1 - e^{2s})\} \quad (54)$$

By using the approximation (42), which is valid for low charge densities, and neglecting $e^{2s} = (a/R)^2$ compared to unity, (52) reduces at low concentrations to

$$\psi_1(a, \lambda) = (kT/\epsilon_2) \ln [e^{2\lambda s}(2 - \lambda)/2(1 - \lambda)^2] \quad (55)$$

and in the further limit of $\lambda \ll 1$ to

$$\psi_1(a, \lambda) = (2\lambda kT/\epsilon_2)(s + 3/4) \quad (56)$$

If we substitute (19) in (56), the limiting potential at the rod is seen to be

$$\psi_1(a, \lambda) = (-2\nu\epsilon_1/hD)(s + 3/4) \quad (57)$$

Equation (53), which corresponds to $\lambda = \Lambda$, reduces at low concentrations to

$$\psi_2(a, \Lambda) = (2kT/\epsilon_2)[s + \ln (-s) + \ln \sqrt{2}] \quad (58)$$

Finally, for $\lambda > \Lambda$, since β^2 decreases rapidly as s and λ increase, we see that (54) has as its limiting form

$$\psi_3(a, \lambda) = (2kT/\epsilon_2)[s - \ln \sqrt{\lambda} + \ln \sqrt{2}] \quad (59)$$

Equations (56), (58) and (59) thus summarize the dependence of $\psi(a)$ on λ and s in the range of low concentrations. At a fixed concentration, $\psi(a)$ increases linearly with λ at first and eventually slows down to a logarithmic increase (s and $-\ln \lambda$ both have the same sign when $\lambda > 1$) at high charge densities. For a fixed charge, the dependence on concentration through $s = \ln (a/R) = (\ln c + \text{const.})$ is *logarithmic* with a coefficient which depends on λ. In other words, at low charge densities (i.e., when $|\epsilon_2\psi/kT| \ll 1$ and the counter ions may readily diffuse away from the rod), the potential (57) at the rod is only a little less than it would have been in the complete absence of counter ions; as the charge increases, the potential at a begins to fall below that of the classical charged rod, because counter ions are now drawn to the rod by electrostatic attraction and $\psi(a)$ increases only as $\ln (\nu/h)$ instead of proportional to ν/h.

The boundary condition (14) shows that the potential initially decreases as r increases from a. The decrease is monotone, because coth x is monotone

and single valued (as is cot x in the range $0 < x < \pi$), and hence the boundary condition (13) establishes $r = R$ as the only point where $d\psi/dr$ vanishes. Since $\psi(a) > 0$, $\exp(-\epsilon_2\psi/kT) > 1$ because ϵ_2 always has the opposite sign from $\psi(a)$; hence $\rho(a) > \bar{\rho}$ for a range of r values near a. Then $\rho(r)$ must be less than $\bar{\rho}$ for some further range of r values because the integral of the density over the whole range of r must satisfy (15). Consequently $\psi(r)$ must vanish for some value r_0, where $a < r_0 < R$, and since $\psi(r)$ is monotone, $\psi(R)$ must have the opposite sign from $\psi(a)$. If the rod has a positive charge (and therefore a positive potential), $\psi(R)$ must then be negative. When $\psi(r)$ vanishes the condition

$$(2\lambda z^2/C)\sinh^2\sqrt{C}\,\xi = 1 \tag{60}$$

must obviously always be satisfied, where

$$z = r_0/R \tag{61}$$

and C and ξ are defined in (8) and (9).

When the charge parameter is small, we may substitute (42) in (22), and obtain

$$\psi_1(R, \lambda) = (-kT/\epsilon_2)\ln\{(1 - \lambda/2)[1 + 2x(1 - \lambda)^2/(2 - \lambda)^2]\} \tag{62}$$

where x is given by (43). For small x and λ, the logarithms may be expanded, giving

$$\psi_1(R, \lambda) = kT\lambda/2\epsilon_2 - (2kTx/\epsilon_2)(1 - \lambda)^2/(2 - \lambda)^2 \tag{63}$$

which has as its limit

$$\psi_1(R, \lambda) = -\nu\epsilon_1/2hD - kTx/2\epsilon_2 \tag{64}$$

It will be noted that the value of $\psi_1(R)$ is always very much smaller in absolute value than $\psi_1(a)$; the ratio is $1/(4s + 3)$. When $\lambda = \Lambda$, (38) immediately gives

$$\psi_2(R, \Lambda) = (kT/\epsilon_2)\ln 2\Lambda \tag{65}$$

which is again far smaller in magnitude than $\psi_2(a, \Lambda)$. For $\beta s = \pi/2$, all the functions assume a simple form, because the transcendental term in (35) vanishes and $\lambda = L$ is given by (46) explicitly in terms of s. Here

$$\psi_3(a, L) = (kT/\epsilon_2)\ln(8s^2e^{2s}/\pi^2) \tag{66}$$

$$\approx (2kT/\epsilon_2)[s + \ln(-s)] \tag{67}$$

and

$$\psi_3(R, L) = (kT/\epsilon_2)\ln 2 \tag{68}$$

Finally, when λ gets very large compared to L, β approaches π/s and (24) becomes

$$\psi_3(R, \lambda) \sim (kT/\epsilon_2)\ln 2\lambda - \pi^2kT/\epsilon_2 s^2 \tag{69}$$

The results of the analysis above show first that $|\psi(a)| > |\psi(R)|$, and second that $\psi(R)$ approaches a value dependent only on λ.

The general course of $\psi(r)$ is now clear: at $r = a$, it has a large positive value, and at some distance r_0 becomes zero, after which it becomes asymptotic to a small negative value. The dependence of z on the parameters will now be discussed. For $\lambda = \Lambda$, the value of $z = r_0/R$ is, of course, easy to determine; in this case, $z\sqrt{2\Lambda}(1 - \ln z)$ must equal unity: for $\Lambda \approx 1$, the root is $z = 0.34$. For other values of λ, approximation methods must be used. Equation (15) provides a convenient starting point: if we let $\psi = \psi(r)$ for the range $a \leq r \leq r_0$ and $\psi = \psi(R)$ for $r_0 < r \leq R$, we are using the correct potential up to r_0, and approximating by a step function the potential in the range beyond r_0. We may use the approximation with confidence, because we have seen that $|\psi(a)|$ is always larger than the asymptotic value $|\psi(R)|$ approached by ψ beyond r_0. To this approximation, (15) becomes for the range $\lambda > \Lambda$

$$z^2[2\lambda - (1 + \beta^2)] = (1 + \beta^2)^2(1 - z^2)^2/4 \qquad (70)$$

if we substitute (18) and (60) for the transcendental terms. Substitution of (35) replaces the coefficient of z^2 on the left of (70) by $\lambda(1 + \beta \cot \beta s)$. Now for large values of λ and s, $(\beta \cot \beta s)$ approaches unity, and β approaches π/s; hence (70) approaches the form

$$A\sqrt{\lambda}z = B(1 - z^2)/2 \qquad (71)$$

where A and B are between one and $\sqrt{2}$. The asymptotic solution of the quadratic for large λ is

$$z \sim 1/2\sqrt{\lambda} \qquad (72)$$

The result shows that the point where the potential drops to zero moves closer to the rod as λ increases, as one might expect; the significant fact is the rate.

The results of our analysis may now be qualitatively summarized. Suppose we allow the charge on the rod to build up from zero to a large positive value, and maintain electrical neutrality by constantly supplying an equivalent amount of counter ions (for instance, by the hypothetical experiment mentioned in the introduction). For low charges, the potential on the rod will be nearly the same as if the counter ions were not present, and the distribution of the latter will be nearly uniform; there will be, however, a slightly higher average concentration of counter ions near the rod and a slightly lower concentration near $r = R$ than would obtain if the rod were not present. In this range of charge density on the rod $|\epsilon\psi/kT| < 1$ everywhere. As the charge parameter increases toward values where $|\epsilon\psi/kT|$ near the rod becomes unity and greater, a strong tendency of the

ions to cluster in the vicinity of the rod appears; as a consequence, the concentration at large distances is diminished below the average concentration corresponding to (an impossible) uniform distribution. In the limit of very high charge densities, the number of counter ions in the space charge near the rod accounts for a large fraction of the total, and the concentration at large distances increases very much more slowly than proportional to the total charge.

Now we allow the radius of the container to increase. Due to the increased volume available, the ionic density near the rod decreases but again only logarithmically due to the electrostatic attraction of the rod, while simultaneously the relative concentration at large distances increases.

These conclusions have a definite bearing on the general problem of polyelectrolytes. The shape of flexible polyelectrolyte molecules is the result of the interaction of Brownian contraction and electrostatic forces. An exact knowledge of the electrostatic potential of rod-like molecules therefore permits in principle the determination of the most probable configuration. The application of a suitable charging process leads to the evaluation of the free energy and thence to the other thermodynamic properties of dilute solutions of polyelectrolytes.

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[1] Verwey, E. J. W., and Overbeek, J. T. G., *Theory of the Stability of Lyophobic Colloids*, Elsevier, New York, 1948; cf. also Loeb, A. L., *J. Colloid Sci.*, **6**, 75 (1951) and Morawetz, H., Thesis, Polytechnic Institute of Brooklyn, 1950.

[2] Katchalsky, A., Künzle, O., and Kuhn, W., *J. Polymer Sci.*, **5**, 283 (1950); Fuoss, R. M., and Strauss, U. P., *Ann. N. Y. Acad. Sci.*, **51**, 836 (1949).

[3] Katchalsky, A., and Lifson, S., unpublished work.

[4] Morawetz (*loc. cit.*) has treated this problem, but his results are not available in the literature at this time.

THE EFFECT OF ULTRA-VIOLET RADIATION ON A RING CHROMOSOME IN ZEA MAYS*

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The analysis of the genetic effects of ultra-violet radiation has progressed farthest in *Zea mays*. As a result of the work of Stadler and his colleagues it has become apparent that while mutations are produced in quantity by the treatment, chromosome rearrangements, so ordinary following x-ray treatment, are quite rare (Stadler, 1941) Moreover, the chromosome breaks found appear to be preponderantly single breaks, a circumstance also found cytologically by Swanson in Tradescantia (Swanson, 1942). The issues raised here are the relation of the mutation process to the breakage of chromosomes, or stated in a different way, the relation of the linear order of the genes to their stability. The existence of a mutagenic agent producing chromosome breaks with a low frequency would imply that the two processes are dissociable. The experimental difficulty arises in the detection of chromosome breaks. Following a disturbance at any point in the chromosome, the restitution of the linear linkage may not occur (single break), may occur in a changed sequence (rearrangement) or may reestablish the original connection (restitution). The restituted breaks are not detectable in the usual experiments; it therefore remains possible that a mutagen may fail to produce rearrangements, yet have a major effect on disruption of the linear order, an effect which might be of importance in considering the nature of the mutation process. In the present paper experiments will be presented in which it is shown that ultra-violet radiation of maize pollen carrying a ring chromosome leads to loss of the ring with a high frequency. This result is interpretable as showing either that breakage and restitution are one of the most frequent effects of the radiation, or that some other phenomenon affected (e.g., inhibition of gene reproduction) may have as its consequence the formation of fragmented chromosomes.

Since the work of L. V. Morgan on Drosophila (1933) and McClintock on maize (1932, 1938), it has been clear that ring chromosomes provide a most sensitive material for studying the details of chromosome behavior. Reproduction in a ring chromosome must take place in a single plane; otherwise the daughters will be either interlocked or form a double ring with two centromeres. McClintock (1938) has studied in maize the behavior of both large and small ring chromosomes which provides the basis of variegation in plants that contain them. The small rings form doubles that are

usually lost at mitosis. In the case of the large rings the dicentrics are broken when the cell plate is formed; the daughters being of unequal size in many cases. Of great importance in the present connection, no evidence of the production of rod from ring chromosomes was found. It is clear that this material provides a useful test of the supposed behavior of ultra-violet radiation in producing single breaks—which would make rods out of rings.

TABLE 1

EFFECTS OF RADIATION ON A^b RING CHROMOSOME *Dp 3a* IN ENDOSPERM

A. Functional Pollen: *a x-3*/Dp 3*a*

Control	NO. OF EARS	N	VARIEGATED A	FRACTIONAL A	*a*
I	12	352	85.8 ± 1.8	8.5 ± 1.4	5.7 ± 1.2
II	1	41	36.5 ± 7.4	9.7 ± 4.6	(53.7)
III	1	41	73.2 ± 6.9	26.8 ± 69	...
2967 Å, 6 sec					
I	9	396	71.0 ± 2.3	14.1 ± 1.7	12.37± 1.7
2967 Å, 30 sec.					
I	3	99	52.5 ± 5.0	24.2 ± 4.3	22.2 ± 4.1
2967 Å, 120 sec.					
I	16	634	45.0 ± 2.0	37.0 ± 1.9	18.0 ± 1.5
II	5	263	29.3 ± 2.8	30.8 ± 2.8	39.9 ± 3.0
III	1	48	58.4 ± 7.1	18.7 ± 5.6	22.9 ± 6.1
2537 Å, 3 sec.					
I	7	285	53.7 ± 2.9	28.1 ± 3.1	18.2 ± 2.3
II	1	42	23.8 ± 6.5	38.1 ± 7.4	38.1 ± 7.4
III	1	23	56.5 ± 10.2	30.4 ± 8.4	13.0 ± 7.0
4 inch					
I	1	30	66.7 ± 8.6	23.3 ± 7.7	10.0 ± 5.5
III	2	102	35.3 ± 4.7	33.3 ± 4.7	31.4 ± 4.6
X-rays, 1000 r					
I	8	203	82.8 ± 2.6	7.9 ± 1.9	9.3 ± 2.0
1000 r + 2967 Å, 30 sec.	6	219	48.4 ± 3.4	25.1 ± 2.9	26.5 ± 2.9

I B. Functional Pollen: *a*/Dp 3*a*; *a x-3*/Dp 3*a*

Control	3	221	46.6 ± 3.3	2.26 ± 1.4	50.2 ± 3.4
2537 Å, 3 sec.	4	159	23.2 ± 3.3	16.4 ± 2.9	60.4 ± 3.8
4 inch	2	38	55.3 ± 8.1	21.1 ± 6.7	23.6 ± 6.8
X-rays, 1000 r	2	129	35.7 ± 4.2	2.3 ± 1.3	62.0 ± 4.8

The rod chromosomes would be stable and hence any effect of the radiation in producing them would be detectable by failure of variegation to occur where ordinarily it would be expected. The critical test in maize must be made in the plant where according to McClintock's (1941) work broken chromosomes are stabilized.

Plan of the Experiments.—The experiments were possible because of the preparation, in connection with the study of a group of x-ray induced muta-

tions at the *A* locus (Stadler and Roman, 1948), of a series of stocks containing a small duplication (Dp 3*a*) for the region of chromosome 3 in maize including the A^b allele. This duplication shows variegation of the type expected from ring chromosomes for the color affected by the *A* gene, both in the plant and the endosperm. According to Stadler and Roman, it usually appears as a collapsed ring at pachytene; root tip smears have shown it, in connection with the present experiments, as a definite ring in the meristematic divisions.

Dr. Stadler supplied the following series of crosses:

Experiment I (field crop)	Experiment B (greenhouse)
a B Pl × *a-X3*/Dp 3*a*	*a B Pl* × *a*/*a-X3*/Dp 3*a*
a pr su × *a-X3*/Dp 3*a*	Experiment II and III (greenhouse)
a-X1/a^p (or *a*) × *a-X3*/Dp 3*a*	*a-X2* (or *a-X4*)/a^p (and *a*)

Two series of experiments were carried out, experiment I (see tables 1 and 2) consisting of a group of field-grown plants and an earlier experiment in the greenhouse, listed in the same tables as experiments II and III. The male parent in all these cases was *a-X3*/*a-X3*/Dp 3*a*, so that the only functional pollen carried the ring. The experiment listed as B came from a male parent of the constitution *a*/*a-X3*/Dp 3*a*, half the functional pollen

TABLE 2

FREQUENCY OF TYPES OF KERNELS SHOWING AREAS OF DEGENERATE ENDOSPERM, IN MATINGS OF *a*/*a-X* 3 × *a-X3*/Dp 3*a*

	VARIEGATED	FRACTIONAL	*a*	GERMLESS	N
Control					
I	96.9 ± 2.2	3.1 ± 2.2	...	...	61
II	100	...	...	...	14
III	84.1 ± 4.6	15.9 ± 4.6	...	3.1 ± 2.1	63
2967 Å, 6 sec.					
I	93.9 ± 2.5	6.1 ± 2.5	...	2.0 ± 2.	49
2967 Å, 120 sec.					
I	44.0 ± 3.4	66.0 ± 3.4	...	12.0 ± 6.7	25
II + III	53.3 ± 4.1	14.7 ± 2.9	32.0 ± 3.8	12.6 ± 2.7	150
2537 Å, 4 sec.					
III	57.6 ± 6.1	6.1 ± 2.9	36.3 ± 5.9	13.6 ± 4.2	66
X-rays, 1000 r					
I	84.6 ± 3.8	15.4 ± 3.8	...	57.1 ± 5.1	91

being *a* without the duplication. In these crosses the colorless kernels came from two sources, from the gametes with the ring loss and from those never having received the ring.

The three types of female parents allow three different types of test. From the *a B Pl* parents, comparisons of the ring behavior in plant and endosperm could be obtained: colorless patches in the kernel, and the absence of anthocyanin in the plant with the full complement of color genes

other than *a*. The pollinations on the *a pr su* females gave tests in the endosperm simultaneously for the *A* locus in the ring and for the *Pr* and *Su* loci in the rod chromosomes. Finally, the tests with the genotypes carrying the deficient chromosomes gave information about the behavior of the other loci included in the ring, rather closely linked to *A*, but showing a characteristic degenerate endosperm, and giving rise to embryo abortion in the homozygous deficiency.

Two wave-length regions were used for the treatment, 2537 Å (from an Hg resonance lamp) and 2967 Å, and longer wave-lengths isolated from the mercury arc spectrum by a diphenyl filter referred to here as 2967 Å. Dosage was varied by changing the time of exposure. For the details of the treatments, the paper of Stadler and Uber (1942) should be consulted.

For the comparison of the ultra-violet data with x-ray effects, a dosage of 1000 r units was used. This experiment was complemented by one in which following the x-ray treatment, a 30 sec. exposure of ultra-violet light at wave-length 2967 Å was applied.

Experimental Results.—The endosperms of kernels carrying A^b in the ring may be grouped into three classes. The first, and the most frequent in untreated material, is the class showing small sectors of colorless aleurone scattered over the surface, when *a* is the character being classified. In plants of the constitution *a-X/a-X*/Dp 3*a*, these small sectors are degenerate, and show as small pitted areas. This variegated class is easily distinguishable from the second class, the fractionals, in which half or more of the kernel is either colorless or degenerate, and the remainder variegated in the usual ring pattern. This fractional pattern is like that already discussed by Stadler in the progenies from irradiated rod chromosomes. In the present classification, kernels with even a small spot of color (like the "recovery" spots) are included in this group. Finally there are the completely colorless or degenerate seeds.

The endosperm data for three different experiments in which the functional pollen was of the constitution *a-X3*/Dp 3*a*, are shown in table 1, A. Experiment I consists of data from three different pollen parents, all of the constitution denoted, crossed to *a-X1/a* or *a; a pr su;* and *a B Pl* plants. Tests for homogeneity using the X^2 method gave a value of *P* between 0.50 and 0.30, hence it was considered legitimate to combine the data. In experiment II, the cross of *a-X3/a* in the greenhouse series, the single control ear showed poor color development in part of the ear, giving a group of seeds faintly colored, classified in the table as colorless. Plant tests on these seeds showed one green seedling out of the 13 tests, indicating that most of the seeds had carried the ring. Experiment III differed in that, as already indicated a^p was present, instead of the *a* allele in II, but was also part of a greenhouse crop. In table 1, only the *a* or a^p/*a-X3* seeds are recorded; the *a-X3/a-X3* (or *a-X1*) seeds are found in table 2.

The frequencies differ in the different experiments, in some cases significantly: for example, compare the values for the 120 sec. exposure at 2967 Å, in experiments I and II. These differences are not surprising, considering the conditions of the two experiments: greenhouse vs. field crop, possible fluctuations in exposure, etc. (cf. Stadler and Uber). The important point is that all agree in the order of magnitude of the effect, a sharp increase in the frequency of the fractional and colorless seeds at the expense of the variegated type. This increase holds for all the ultra-violet exposures, at both the wave-lengths used. In experiment I, intermediate dosages at 2967 Å indicate a leveling off at the 30 sec. exposure; but further data are obviously necessary to define the shape of the dosage effect curve. Similarly, for the 2537 Å exposure, the 3 and 4 second groups are very similar in effect. More detailed dosage-effect curves are needed before discussion of the relative effectiveness of the two wave-lengths.

Table 1, B gives data from crosses in which half the seeds are expected to be colorless (*aa*), deriving from pollen without the ring. In the 4 sec. exposure, the frequency is lower than expected on this basis; but the explanation probably lies in the possibility that two rings were present in the particular pollen parent used for this treatment. The data corroborate those of the more clear-cut cross. The frequency of fractional kernels rises, and the increase in colorless seeds is also perceptible.

The effect of x-rays on *A*-loss in the ring chromosome was only studied at one dose, 1000 r units, fairly heavy, producing 12.3% germless seed in this experiment. There is an increase in the percentage of colorless seeds, but the fractionals remain unchanged. The result is in accordance with other x-ray data: the production of fractionals by x-ray treatment is relatively low, compared to that in the ultra-violet. In the 120 sec. ultra-violet exposure, in which the frequency of germless seeds was less than 1%, the fractionals reach 37% (experiment I). These relationships resemble those already found in the study of rod chromosomes (Stadler, 1941).

Since the ultra-violet effects are of such a magnitude, it seemed worthwhile to test the combination of x-ray and ultra-violet treatment. Swanson's work on Tradescantia (1944) and Kaufmann and Hollaender's work on Drosophila (1946) had shown an inhibitory effect of post-treatment with ultra-violet light on the formation of chromosome rearrangements by x-rays. Since the effects of the two types of radiation on the ring chromosome differ, it should be possible to see effects of the x-ray treatment on the action of the ultra-violet light in the endosperm from treated pollen. Comparison of the treatment (Table 1 A) with 30-inch 2967 Å alone, and with a pretreatment with 1000 r of x-rays, shows that the ultra-violet effect is the same in both groups. The x-ray effect, as far as could be judged by the frequency of germless seeds (none were found in the doubly treated group with an expectation of twenty), was inhibited.

The classifications of the seeds showing degenerate endosperm patches, from the proper crosses in experiment I and from experiments II and III, are given in table 2. Again the pronounced effect of the ultra-violet treatment is seen, on the production of fractionals. The completely colorless seeds were seen only in the greenhouse crop, and may have had small areas of colored aleurone. In any case, from these experiments it is apparent that the effect of the ultra-violet is not on the *A* gene alone in the ring, but involves the other loci as well, in the various *a-X* deficiencies. Following the interpretation of Stadler and Roman, the data of table 2 indicate that the ultra-violet treatment causes loss either partial or complete of the Dp *3a* ring chromosome. One additional fact appears in table 2. The homozygous deficiency does not survive in the plant; hence, embryos from which the ring chromosome had been eliminated at an early stage would not survive. Such seed would be germless, and an effect of the ultra-violet on the ring loss in the embryo should be detected as an increase in the percentage of germless seed. This does in fact occur, agreeing with the tests of the next section.

Frequency of Loss of Dp 3a in the Embryo.—Seeds from

TABLE 3

FREQUENCY OF GREEN SEEDLINGS IN PROGENIES FROM POLLEN CARRYING THE RING DUPLICATION

	TYPE OF SEED															
	VARIEGATED A				FRACTIONAL A								TOTAL			
	a	N	%		*a*	N	%		*a*	N	%		*a*	N	%	
Control																
I	6	149	4.0	± 1.5	1	20	5.0	± 4.8	3	8	...		9	176	5.1	± 1.7
II + III + B (var. only)	2	118	1.7	± 1.2	1	13	7.7		1	13	7.7		4	144	2.8	± 1.4
2967 Å, 6 sec.																
I	11	151	7.2	± 2.1	4	22	18.2	± 8.2	3	24	12.5	± 6.7	18	197	9.1	± 2.0
2967 Å, 30 sec.	4	53	7.5	± 3.8	2	22	9.1	± 6.1	4	22	18.2	± 8.2	10	97	10.3	± 3.1
2967 Å, 120 sec.																
I	48	193	24.9	± 3.1	57	172	33.1	± 3.6	27	71	38.6	± 5.7	132	436	27.5	± 2.1
II	6	56	10.7	± 4.1	9	63	14.3	± 4.4	10	69	14.5	± 4.2	25	188	13.3	± 2.5
2537 Å, 3 sec.	11	72	15.3	± 4.2	10	47	21.2	± 5.9	10	48	20.8	± 5.8	31	167	18.6	± 3.0
II + III + B	10	35	28.6	± 7.6	8	27	29.6	± 8.8	2	9	22.2		20	71	28.2	± 5.3
TOTAL	90	560	16.1	± 1.6	90	353	25.5	± 2.3	56	243	23.0	± 2.7	...	1156	...	
X-rays, 1000 r	11	94	11.7	± 3.3	1	11	9.1		6	12	50		18	117	14.5	± 3.2

the crosses to the *a B Pl* parents, whose progeny would be fully colored plants, were planted and allowed to grow until they could be classified definitely as either full colored, sectorial or green seedlings. The usual behavior of the Dp *3a* chromosome in the plant produces a striping for the anthocyanin color, the areas where the ring has been lost lacking the anthocyanin, due to the *A* loss (Stadler and Roman, 1948). If the ring were transformed into a rod, the condition for this variegation would be lost, and full-colored plants should be found. In table 3, the results of the seedling tests are tabulated. In the total of approximately 1000 plants, not one was full-colored. Thus, if the breaks in the ultra-violet-treated chromosomes occur with any frequency, they behave in such a way as to maintain the ring structure.

In table 3, the seedlings are tabulated according to the classification of the kernels from which they came. This requires no comment for experiment I; to save space, experiments II, III and B have been combined. Since none of the colorless kernels of experiment B were tested, the seedlings from *a* kernels in these entries, therefore, come only from experiments II and III, and the totals differ from those in table 1.

The data show clearly that with the increase in exposure to ultra-violet radiation, the loss of the ring chromosome is greatly increased at both wavelengths. There is also a significant increase following the x-ray treatment, of somewhat greater magnitude than the effect on the endosperm. The frequencies differ in the classes from the three groups of kernels. It is to be expected that the frequency of *a* seedlings from *a* kernels be higher than the mean frequency, since this class includes those cases of ring loss occurring in the pollen-grain divisions, giving endosperm and embryo gametes equally without the ring. A difference of possible significance, however, occurs between the frequency of green seedlings in the variegated and the fractional classes of the 120 sec. 2967 Å treatment of experiment I, a difference of $8.2 \pm 4.7\%$. This indicates, although just below statistical significance, that pollen grains favorably situated are apt to have both sperm nuclei affected, as regards ring-chromosome loss. It should also be noted that the differences in the other treatments are all in the same direction, and the differences between the totals of the treated samples in the variegated and fractional groups show a statistically significant difference. The analysis of Stadler and Uber has shown that both sperm nuclei tend to lie in a similar position in the pollen grain. This spatial factor would lead to a coincidence of effects in both sperm nuclei.

The maximum frequency of ring loss in the embryo judged by frequency of green seedlings is 27.5% in the 120 sec. 2967 Å treatment. This frequency is of the same order of magnitude as the frequency of complete *a* loss in the endosperm (actually higher, in the same experiment). This result is in striking contrast to the rarity of deficiencies in plants from rod chromosomes treated with ultra-violet light (Stadler, 1941).

Ring Chromosomes and the Problem of Chromosome Breakage by Ultra-Violet Light.—It seems legitimate to interpret the data presented in terms of a high frequency of ring-chromosome loss induced by the treatment. It is difficult in the present case to make the quantitative comparison carried out for the x-ray effects in Drosophila, where a rod chromosome of the same size as the ring was available for treatment (Bauer, 1943; Muller, 1950). The best comparison possible at present is by way of the relative frequency of induced deficiency for the *Su* locus, in chromosome 4, to the frequency of *A* loss in ring and rod. *Pr*, unfortunately, was not usable in the present experiment, since the pollen parents were heterozygous for the recessive, and the frequency of complete loss could not be determined. At the 120 sec. exposure of 2967 Å, out of 99 mosaic kernels, 15 were losses of *Su* and 84 were losses of *A* from the ring. In the data of Stadler and Uber (1942), table 5 gives a comparable group, at the dose of 32.9 M. Out of 76 deficient seeds, 22 had lost *Su* and 46 had lost *A*. The ratio of *A* loss to that of *Su* is much higher in the ring than in the normal chromosome. This difference becomes more meaningful when the locus of *A* in the terminal fifth of the normal chromosome is recalled, with the long distance of chromosome between *A* and the centromere available for breakage to produce deficiency. This is a much larger segment of chromosome than is contained in the ring, yet in relation to the *Su* loss, the ring is more frequently affected.

There are two modes of approach to the problem of the high frequency of ring loss. One, for which there is no evidence at present, would be based on the assumption that the centromere is peculiarly sensitive to ultra-violet radiation; in other words "mutates" frequently, is inactivated, and its chromosome lost. A rod chromosome indentical with the ring could provide a test. The hypothesis seems unlikely, but cannot be excluded in our present stage of knowledge.

The more plausible postulate is based on the spontaneous aberration of ring chromosomes, already referred to. The formation of double or of interlocked rings as the result of more than one plane of reproduction ("crossing-over" between sister chromatids) can be visualized as an effect of a treatment inducing either breaks in the chromosome, or localized defects in the reproduction of the ring chromosome. It is clear that in the present ambiguous state of definition of the primary event leading to the formation of a chromosome fragment, the distinction between the two possibilities may merely be verbal. The major difficulty, as is evident from the inspection of such summaries as those of Catcheside (1948), Lea (1947), Kaufmann (1948) or Muller (1950), is that a chromosome reproduction intervenes before the "break" is detected. What is observed is the formation of a fragment, or a rearrangement of the chromosomes. From this point of view, any association between chromosome breakage and mutagenic capacity of a given agent might simply indicate that both phenomena stem from a primary effect on chromosome (or gene) reproduction.

In the ring chromosome, either a defect in reproduction at any point in the ring, or a "break" in the chromosome would lead to two alternatives in the formation of the daughter chromosomes. The linear order might be restored between the original chromatids, or crosswise between the sister chromatids. These alternatives exist also in rod chromosomes, but no distinction is possible. They were originally discussed in terms of the detection of sister-strand crossing-over (L. V. Morgan, 1933) in the ring chromosomes in Drosophila, and the application to the induced breaks is straightforward. The consequences of the radiation are realized in the cell generation following fertilization. A double or interlocked ring could be lost from the cell, giving rise to the colorless kernels and the green seedlings of the present series of experiments. If not lost, as in McClintock's R2 ring (1938), both might be incorporated in one of the daughter cells. In this case, it is conceivable that interlocked rings might be able to form disentangled daughter chromosomes, and that the fractional kernels might come from this source. It may be possible that the change in ratio between fractional and colorless kernels at the different dosages might be interpretable as a change in the frequency with which interlocked rings are formed, these requiring one break more than the dicentrics. In the controls of experiment I, the fractional/colorless ratio is 1.42, varying in the treated group through 1.14, 0.92 and finally, at the high 2900 Å dosage, 2.13.

The inhibitory effect of the ultra-violet on chromosome rearrangements from x-rays has been interpreted by Swanson as an immobilization of ends due to coagulation of a matrical substance, favoring restitution. The same explanation is needed to account for the failure of rearrangements to occur in the ultra-violet, if the present conclusions on the basis of ring loss are to be generalized. Ultra-violet radiation then inhibits the formation of rearrangements from the breaks it itself produces.

On the assumption that the loss of the ring is due to sister-chromatid restitution, the calculation of the total effect becomes of interest, as in the x-ray analysis (Bauer, 1943; Muller, 1950). The total effect should then be equal to twice the sum of the fractional plus the colorless kernels in the endosperm. At the 30-inch treatment with 2967 Å, this value comes to 2×46.4, or 92.8%. In other words, almost all the nuclei have been hit at this dose. In the plant, the fractionals are not detected; but as already pointed out, the values for the green seedlings in the plant are comparable to those for the colorless seeds. If it is permissible to extend the reasoning to normal rod chromosomes, the major effect of ultra-violet radiation on chromosomes would seem to be damage to the maintenance of linear order. In view of the fact that the chromosome effects of the different wave-lengths of ultra-violet have been found where studied to follow the absorption spectrum of nucleic acid (see Stadler and Uber, *loc. cit.*), it will be interesting to know whether this is also true for the ring loss.

Summary.—1. The effects of ultra-violet and of x-radiation on a ring-shaped chromosome in *Zea mays* (Dp 3*a*, carrying the A^b locus) have been studied.

2. In the endosperm, it is calculated that at the upper dose given, practically all of the ring chromosomes were affected by the ultra-violet radiation, although a dose of x-rays producing a more severe effect on embryo abortion has a much lower effect on the ring loss.

3. A similar high frequency of ring loss is found in the sporophyte. The effect can be interpreted to mean that the restitution of disruptions of linear order in the chromosome takes place, and can be detected in the ring. It would follow that the low frequency of broken chromosomes found in plants from irradiated pollen, as compared with the endosperm, is due to a higher rate of restitution in the plant.

4. A combined treatment of x-rays with ultra-violet showed no change of the ultra-violet effect by the x-rays, although the usual inhibition of the x-ray effect by the ultra-violet treatment was found.

* This work was begun in 1943 at the University of Missouri, where the radiation treatments were carried out and the plants grown. Some of the seedling tests were carried out at the Department of Genetics of the Carnegie Institution at Cold Spring Harbor; it is a pleasure to thank Dr. Barbara McClintock for help at that time. Without the enthusiasm of Dr. L. J. Stadler these experiments would not have been undertaken: I am indebted to him not only for the genetic stocks used, but for arranging for the treatment and the growing of the plants. The work has also been aided by an Institutional Grant to the Institute for Cancer Research by the American Cancer Society.

Bauer, Hans, "Röntgenauslosung von Chromosomen—Mutationen bei Drosophila melanogaster. II. Die Haufigkeit des primären Bruchereignisses nach Untersuchungen am Ring-X Chromosom," *Chromosoma*, **2**, 407–458 (1943).

Catcheside, D. G., "Genetic Effects of Radiations," *Advances in Genetics*, **2**, 271–349 (1948).

Kaufmann, B. P., "Radiation Induced Chromosome Aberrations," Brookhaven Conference Report. BNLC-4, 27–35 (1948).

Kaufmann, B. P., and Hollaender, A., "Modification of the Frequency of Chromosomal Rearrangements Induced by X-Rays in Drosophila. II. Use of Ultra-Violet Irradiation," *Genetics*, **31**, 368–376 (1946).

Lea, D. E., *Actions of Radiation on Living Cells*, The Macmillan Company, New York, 1947, 402 pp.

Muller, H. J., "Some Present Problems in the Genetic Effects of Radiation," *J. Cell. Comp. Physiol.*, **35**, Suppl. 1, 9–70 (1950).

McClintock, Barbara, "A Correlation of Ring-Shaped Chromosomes with Variegation in *Zea Mays*," PROC. NATL. ACAD. SCI., **18**, 677–681 (1932).

McClintock, Barbara, "The Production of Homozygous Deficient Tissues with Mutant Characteristics by Means of the Aberrant Mitotic Behavior of Ring-Shaped Chromosomes," *Genetics*, **23**, 315–376 (1938).

McClintock, Barbara, "The Stability of Broken Ends of Chromosomes in *Zea Mays*," *Ibid.*, **26**, 234–282 (1941).

Morgan, L. V., "A Closed X Chromosome in Drosophila melanogaster," *Ibid.*, **18**, 250–283.

Stadler, L. J., "The Comparison of Ultra-Violet and X-ray Effects on Mutation," *Cold Spring Harbor Symp. Quant. Biol.*, **9**, 168–177 (1941).

Stadler, L. J., and Uber, Fred M., "Genetic Effects of Ultra-Violet Radiation in Maize. IV. Comparison of Monochromata Radiations," *Genetics*, **27**, 84–118 (1942).

Stadler, L. J., and Roman, Herschel, "The Effect of X-rays upon Mutation of the Gene A in Maize," *Ibid.*, **33**, 273–303 (1948).

Swanson, C. P., "The Effects of Ultra-Violet and X-Ray Treatment on the Pollen Tube Chromosomes of Tradescantia," *Ibid.*, **27**, 491–503 (1942).

Swanson, C. P., "X-ray and Ultra-Violet Studies on Pollen Tube Chromosomes. I. The Effect of Ultra-Violet (2537 Å) on X-Ray Induced Chromosome Aberrations," *Ibid.*, **29**, 61–68 (1944).

SOUND WAVES IN THE ATMOSPHERE GENERATED BY A SMALL EARTHQUAKE†

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In previous papers[1-5] theoretical and experimental results on the coupling of atmospheric compressional waves to various types of surface waves in the underlying earth or ocean have been presented. Recently Benioff[6] presented a paper describing a remarkable instance of atmospheric waves received at Pasadena from the earthquake of January 24, 1951, 07-17-01, magnitude 5.6, 33° 07′N., 115° 34′W., $\Delta = 265$ km. The microbarograph recorded a train of waves which commenced gradually at about 23:34:00 P.S.T. with periods of about $^3/_4$ second and ended at about 23:39:00 P.S.T. with periods of about 1 sec. (Fig. 1). Benioff had noticed a similar disburbance following a small tremor several years earlier.

About 55 km. of path, starting from the epicenter in the Imperial Valley, crossed valley sediments before reaching intrusive rocks of the mountains. For the valley part of the path we may expect, by analogy with the results of a study of air coupling to Rayleigh waves in the weathered layer in northeast Texas, that the group and phase velocities are given by curves similar to the dimensionless curves in figure 2.[4-5] These curves are only useful for an "order of magnitude" calculation when applied to the Imperial Valley.

In this figure the heavy lines represent the dimensionless phase C/β, and group U/β, velocity curves of Rayleigh waves propagated through an earth whose layering is described below, neglecting coupling to the atmosphere. The dashed lines represent the corresponding curves with the inclusion of coupling to atmospheric compressional waves. The constants used in computing these curves are as follows:

$$\alpha_0 = 1070 \text{ ft./sec.} \qquad \beta_1 = 800 \text{ ft./sec.} \qquad \alpha_1 = \sqrt{3}\beta, \qquad \alpha_2 = \sqrt{3}\beta_2$$
$$\rho_2/\rho_1 = 1.39 \qquad \rho_0/\rho_1 = 0.001 \qquad \mu_2/\mu_1 = 13.77$$

where α is compressional wave velocity, β is shear wave velocity, ρ is density, μ is shear modulus and the subscripts 0, 1, 2 refer respectively to air, surface unconsolidated sediments and bedrock. These values were chosen as representative of near surface conditions often encountered in seismic prospecting.

The group velocity curve in figure 2 is divided into branches I, II and III, each of which represents a different train of waves. Branch I represents the ordinary dispersive Rayleigh waves. These waves first appear as long period oscillations, traveling with the speed of Rayleigh waves in the bottom layer, $U = 0.9194\ \beta_2$. Succeeding waves gradually increase in period, since the group velocity decreases as KH increases. Waves represented by branch I predominate if the source of waves and the detector

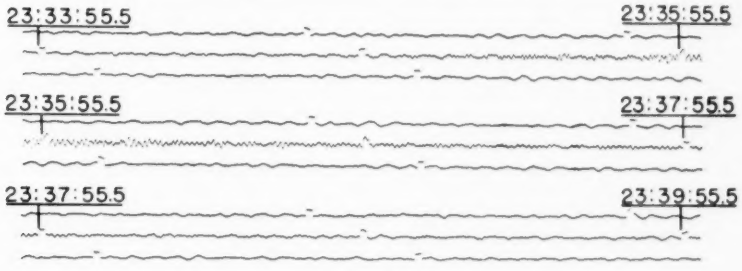

FIGURE 1

Copy of a 35-mm. film record taken with Benioff electromagnetic microbarograph, galvanometer period 0.15 sec., recording speed 0.25 mm./sec.

are located in the earth. Branch II represents a dispersive train of waves beginning with high-frequency oscillations propagated with the speed of Rayleigh waves in the surface layer. The frequency of these waves decreases as time progresses until the waves of both branches have a common frequency, forming an Airy phase[7] which is propagated at the minimum group velocity. Branch III represents an additional train introduced by coupling of the Rayleigh wave to atmospheric compressional waves. The beginning of this train is propagated with the speed of sound in air, and includes all frequencies from zero to that given by KH approximately equal to 2. In the neighborhood of KH = 2, the group velocity curve drops steeply, giving a long train of waves with almost constant frequency, and with phase velocity nearly equal to the speed of sound in air. Branch III governs transmission when the source is in the earth and the detector is in the air, or vice versa.

From figure 2 the depth of the weathered layer required to produce air coupling at a period of 0.9 sec. may be calculated as approximately 100 meters, using the equation

$$H = T(KH)(C/\beta_1)\beta_1/2\pi$$

A more complete discussion has been given previously.[4,5]

An approximate interpretation of the propagation of sound to the microbarograph is that the Rayleigh waves of period 0.9 sec. crossed the valley at a speed of 0.10 km./sec. radiating compressional waves into the air as they went. The time required for these Rayleigh waves to cross the valley was about 550 sec., after which time no Rayleigh waves of sufficiently slow speed to radiate into the air could exist. (The lowest speed is that of the minimum group velocity.) Thus the first airborne waves started for Pasadena at about 07-17-01 G.C.T., a distance of 265 km., while the last ones started at about 07-26-11, a distance of 210 km. From the results of Crary[8] we may approximately consider that these sound waves go vertically to a height of 40 km. with a mean velocity of 0.305 km./sec. and travel the horizontal distance to Pasadena at a speed of 0.326 km./sec. before descending. One can roughly calculate the arrival times as follows:

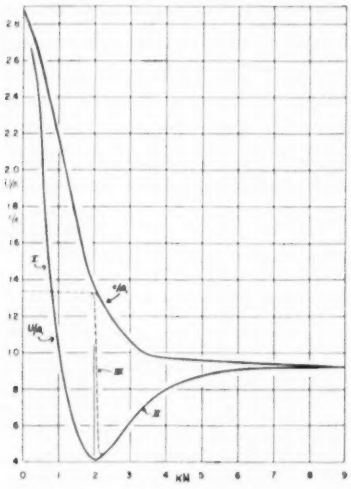

FIGURE 2

Phase and group velocity curves for air coupled Rayleigh waves.

FIRST WAVE	LAST WAVE
Vertical travel time: $2 \times 40/0.305 = 262$ sec.	$2 \times 40/0.305 = 262$ sec.
Horizontal travel time: $265/0.326 = 813$ sec.	$210/0.326 = 644$ sec.
Total travel time: 17 min. 55 sec.	15 min. 6 sec.
Origin time in air: 07-17-01	07-26-11
Computed arrival time: 07-34-56	07-41-17
Observed arrival time: 07-34-00	07-39-00

The values deduced for arrival time of the first and last waves by this order of magnitude calculation agree reasonably well with the observed values in view of the gradual nature of the beginning and end of the signal, the uncertainty of the elastic constants used in the theoretical curves and the simplification of the aerial path.

As mentioned above, the periods within the train are erratic, but show a predominant value of about $^3/_4$ sec. at the beginning and 1 sec. at the end. The most probable explanation of this change is a variation in the dispersion properties of the sediments across the valley.

Regarding the depth of 100 meters, chosen to represent the "weathered layer," it must be pointed out that the Rayleigh wave velocity is essentially controlled by the vertical variation in shear velocity, which may be quite different from that of compressional wave velocities. In a general way the former depends on the degree of lithification and the latter on the water table.

These results illustrate a fundamental reciprocity principle of observation of air-coupled waves through the fact that the Pasadena seismographs did not register this wave train. For a disturbance in either medium, the air-coupled waves are detected in the other medium. Thus a disturbance in the air could produce a similar train of waves in the earth.

This example is believed to represent the first analyzed case of air-coupled waves from a natural earthquake. The conditions necessary for its occurrence are not uncommon and one may expect that additional data will be forthcoming.

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[1] Press, Frank, and Ewing, Maurice, *J. Applied Phys.* **22,** 892–899 (1951).

[2] Press, Frank, Crary, A. P., Oliver, Jack, and Katz, Samuel, *Trans. Am. Geophys. Union,* **32,** 166–172 (1951).

[3] Ewing, Maurice, and Press, Frank, "Tide Gauge Disturbances from the Great Eruption of Krakatoa," paper read at 1951 meeting, Seism. Soc. America.

[4] Press, Frank, and Ewing, Maurice, *Geophysics,* **16,** 416–430 (1951).

[5] Jardetzky, W. S., and Press, Frank, "Rayleigh Wave Coupling to Atmospheric Compressional Waves," *Bull. Seism. Soc. Am.,* in press.

[6] Benioff, Hugo, "Sound Waves in the Atmosphere Generated by a Small Earthquake," paper read at 1951 meeting, Seism. Soc. America.

[7] Pekeris, C. L., Memoir 27, Geol. Soc. America (1948).

[8] Crary, A. P., Geophysical Research Paper No. 5, Air Force Cambridge Research Laboratories (1950).

ON FINITE GROUPS WITH TWO INDEPENDENT GENERATORS. I

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1. Introduction.—Although the theory of finite groups is a well-worked branch of mathematics, the present topic in its full generality is—to the author's knowledge—entirely new; so are the many results obtained.

The title denotes a group Γ all of whose elements are expressible in the form A^xB^y where A, B are independent elements of order m, n respectively: $A^m = 1$, $B^n = 1$. "Independent" means that $A^xB^y = A^uB^v$ implies $A^x = A^u$, $B^y = B^v$, i.e., $x \equiv u \pmod m$, $y \equiv v \pmod n$. It is helpful to imagine the mn elements constituting the group to be distributed in a checkerboard array of m files and n rows, x designating the file ($0 \leqq x < m$) and y the row ($0 \leqq y < n$) in which the element A^xB^y is placed.

Of course, abelian groups with a basis of two elements are a particular instance of a group Γ, but practically all the interest resides in the non-commutative case: $BA \neq AB$. Dihedral groups are a very special example: $A^m = 1$, $B^2 = 1$, $BA = A^{-1}B$.

Our problem is to show how to determine all two-generator groups, at least theoretically, and to study their special properties.

In the series beginning with this note we present some of our main results; a more detailed account, with proofs, will be published elsewhere.

2. The Conjugate Substitutions θ, ϕ.—By the closure of the group Γ, functions $\theta(x)$, $\psi(x)$ must exist such that[1]

$$BA^x = A^{\theta(x)}B^{\psi(x)}, \tag{21}$$

also functions $\phi(y)$, $\omega(y)$ such that

$$B^yA = A^{\omega(y)}B^{\phi(y)}. \tag{22}$$

$\theta(x)$ (as well as $\omega(y)$) is determined only up to multiples of m, and must be periodic modulo m with period m—by which we mean: $\theta(x + m) \equiv \theta(x) \pmod m$, or (what is tantamount): $x \equiv y \pmod m$ implies $\theta(x) \equiv \theta(y) \pmod m$.

To obtain a normal form of $\theta(x)$, we may impose the restriction $0 \leqq \theta(x) < m$, and then $\theta(x)$ is strictly periodic with period m. From this normal form the general form of $\theta(x)$ arises by the addition of $mM(x)$ where the function $M(x)$ is arbitrary.

Using the cancellation law of groups, and the independence of A and B, we can prove that $\theta(x) \equiv \theta(y) \pmod m$ *only* of $x \equiv y \pmod m$. For

$\theta(x) \equiv \theta(y) \pmod{m}$ implies successively: $A^{\theta(x)} = A^{\theta(y)}$, $BA^xB^{-\psi(x)} = BA^yB^{-\psi(y)}$ (by (21)), $A^xB^{-\psi(x)} = A^yB^{-\psi(y)}$, $x \equiv y \pmod{m}$.

In short, $x \equiv y \pmod{m}$ is *equivalent* to $\theta(x) \equiv \theta(y) \pmod{m}$, and consequently $\theta(x)$ defines a *permutation* or *substitution* of the residue classes modulo m, or of their representative elements $0, 1, 2, \ldots, m-1$. We shall refer to this system of integers as $[m]$ or as the *range* m. Obviously $\theta(0) \equiv 0 \pmod{m}$, so that the substitution θ *always leaves* 0 *fixed*. Similarly, ϕ defines a 0-fixing substitution on the range $n = 0, 1, 2, \ldots, n-1 = [n]$.[2]

Since, by (21) and the associative law,

$$B \cdot A^xB^y = A^{\theta(x)}B^{\psi(x)+y} \tag{23}$$

we see that left multiplication by B of all the elements of Γ produces the permutation θ on the files of our checkerboard array—whereby, furthermore, consecutive elements in the file x go over into consecutive elements in the file $\theta(x)$. Here "consecutive" is to be interpreted as follows: "y' consecutive to y" means $y' \equiv y + 1 \pmod{n}$, so that, e.g., 0 is consecutive to $n-1$.

Similarly, right multiplication by A produces the permutation ϕ on the rows of Γ, whereby consecutive elements (mod m) in the row y go over into consecutive elements (mod m) in the row $\phi(y)$.

This representation of θ, ϕ as file, row permutation respectively of the checkerboard arrangement of Γ may be taken as their most graphic description.

The substitutions θ, ϕ *determine the structure of* Γ *completely*—in fact, by applying the associative law and the independence of A and B, we find

$$B^yA^x = A^{\theta^y(x)}B^{\phi^x(y)}, \tag{24}$$

so that the product of any two elements of Γ is given by the rule:

$$A^uB^v \cdot A^xB^v = A^{u+\theta^v(x)}B^{v+\phi^x(y)}. \tag{25}$$

It is evident from this that Γ is abelian if and only if θ, ϕ reduce to the identity on their respective ranges.

We shall term θ, ϕ *conjugate* substitutions. The relation is evidently symmetric, since (24) implies by the taking of inverses and interchange of x, y:

$$B^yA^x = A^{\phi^y(x)}B^{\theta^x(y)}, \tag{26}$$

where $A = B^{-1}$, $B = A^{-1}$; so that ϕ, θ correspond as file, row substitution respectively to the same group Γ regarded as generated by B^{-1}, A^{-1}.

We shall find that each substitution θ (or ϕ) of a conjugate pair has *by itself* some very special properties, and shall therefore designate as *special* any substitution associated with a two-generator group Γ as file (or row)

permutation. The sequel revolves about the determination and study of these special substitutions.

As not too trivial an example of a pair of conjugate substitutions, we may cite

$$\theta = (0)(2)(4)(6)(1357), \qquad \phi = (0)(2)(4)(6)(1753), \tag{27}$$

each on the range 8.[3] They define a group Γ of order 64, whose multiplication table can be inferred completely from the following relations (cf. (24)):

$$A^8 = 1, B^8 = 1; \qquad BA = A^3B^7, BA^2 = A^2B^5, BA^3 = A^5B^3, BA^4 = A^4B. \tag{28}$$

For example: $B^2A^5 = B \cdot BA^4 \cdot A = B \cdot A^4B \cdot A = BA^4 \cdot BA = A^4B \cdot A^3B^7 = A^4 \cdot BA^3 \cdot B^7 = A^4 \cdot A^5B^3 \cdot B^7 = AB^2$, agreeing with (24).

We may remark in passing that each of the substitutions θ, ϕ is also self-conjugate.

3. Criteria for Conjugate Special Substitutions.—We begin with

THEOREM I. *In order that two substitutions θ, ϕ on the ranges m, n and of periods r, s, respectively, shall be conjugate special, three conditions are necessary and sufficient, namely:*

$$\begin{aligned}
&\text{(I)} \quad \theta(0) = 0, \qquad \phi(0) = 0 \\
&\text{(II)} \quad s \mid m, \qquad r \mid n \\
&\text{(III)} \quad \theta^y(x + t) - \theta^y(t) \equiv \theta^{\phi^t(y)}(x) \pmod{m} \\
&\qquad\qquad \phi^y(x + t) - \phi^y(t) \equiv \phi^{\theta^t(y)}(x) \pmod{n}
\end{aligned}$$

for all x, y, t.[4]

Condition (III), which guarantees the associative law for Γ, may be lightened in any of the following three ways: by putting $t = 1$, by putting $y = 1$, by putting $t = 1$ in one of the formulas and $y = 1$ in the other. In this way we get, respectively, (III.1), (III.2), (III.3), each of which—in the presence of (I), (II)—implies the full condition (III).

$$\begin{aligned}
&\text{(III.1)} \quad \theta^y(x + 1) - \theta^y(1) \equiv \theta^{\phi(y)}(x) \pmod{m} \\
&\qquad\qquad \phi^y(x + 1) - \phi^y(1) \equiv \phi^{\theta(y)}(x) \pmod{n}. \\
&\text{(III.2)} \quad \theta(x + t) - \theta(t) \equiv \theta^{\phi^t(1)}(x) \pmod{m} \\
&\qquad\qquad \phi(x + t) - \phi(t) \equiv \phi^{\theta^t(1)}(x) \pmod{n}. \\
&\text{(III.3)} \quad \theta(x + t) - \theta(t) \equiv \theta^{\phi^t(1)}(x) \pmod{m} \\
&\qquad\qquad \phi^y(x + 1) - \phi^y(1) \equiv \phi^{\theta(y)}(x) \pmod{n}.
\end{aligned}$$

By a *translation modulo m* we shall understand the change of variables represented by

$$x \equiv x' + a, \qquad y \equiv y' + b \pmod{m} \tag{31}$$

A translation of any 0-fixing substitution $y \equiv \theta(x) \pmod m$ is again a substitution, $y' \equiv \theta(x' + a) - b \pmod m$, which is 0-fixing if and only if $a \equiv t$, $b \equiv \theta(t) \pmod m$. The substitution so defined, namely $x \to \theta(x + t) - \theta(t)$, will be termed the *t-translation* of θ and denoted by $T_t\theta$. Obviously, $t' \equiv t \pmod m$ implies $T_{t'}\theta = T_t\theta$. We easily prove that

$$T_t(T_{t'}\theta) = T_{t+t'}\theta, \tag{32}$$

and also observe $T_0\theta = \theta$.

In terms of translations and powers, the conditions (III), (III.1, 2, 3) may be restated as follows.

(III′) $T_t\theta^y = \theta^{\phi^t(y)}$, $T_t\phi^y = \phi^{\theta^t(y)}$
(III′.1) $T_1\theta^y = \theta^{\phi(y)}$, $T_1\phi^y = \phi^{\theta(y)}$
(III′.2) $T_t\theta = \theta^{\phi^t(1)}$, $T_t\phi = \phi^{\theta^t(1)}$
(III′.3) $T_t\theta = \theta^{\phi^t(1)}$, $T_1\phi^y = \phi^{\theta(y)}$.

4. Criteria for a Special Substitution. Derivative.—THEOREM II. *In order that a 0-fixing substitution θ on the range m and of period r be special, it is necessary and sufficient that a function $\psi(t)$ on $[m]$ to $[r]$ shall exist such that*

$$T_t\theta = \theta^{\psi(t)} \tag{41}$$

—in other words, every 0-fixing translation of θ shall be a power of θ.

THEOREM III. *In order that a 0-fixing substitution θ on the range m and of period r be special, it is necessary and sufficient that there should exist a corresponding substitution θ_1 on the range r such that, for all y,*

$$T_1\theta^y = \theta^{\theta_1(y)}. \tag{42}$$

In other words, the 1-translation of every power of θ shall be again a power of θ; the function $\theta_1(y)$ on $[r]$ to $[r]$ so defined will then of necessity be a substitution. θ_1 is evidently unique, if at all existent; it will be called the *derivative* of θ.

Example: Each of the substitutions θ, ϕ specified in (27) has the derivative $\theta_1 = \phi_1 = (0)(2)(13)$.

THEOREM IV. *The derivative θ_1 of any special substitution θ is itself special—in fact, θ_1 is a conjugate of θ.*

θ_1 may be regarded as the principal conjugate of θ; it is the unique conjugate whose range is equal to the period r of θ. This is the minimum range of a conjugate of θ; any other conjugate—if at all existent—is on a range whose extent is a multiple vr of r, where $v > 1$.

The group defined by a special substitution and its derivative: θ, θ_1, will be termed a *primary group*. In a sense, this kind is the most fundamental, since the general group Γ can be constructed by proper pairing of primary groups (cf. the paragraph preceding Theorem VI).

THEOREM V. *The derivatives θ_1, ϕ_1 of conjugate substitutions θ, ϕ are themselves conjugate.*

By repetition, this conclusion applies to the derivatives θ_i, ϕ_i of any order i, as defined in the next section.

5. Successive Derivatives.—Since θ_1 is special, it has a derivative θ_2, which is special, and therefore has a derivative θ_3, etc. In this way, we construct the series of derivatives

$$\theta_0 = \theta, \theta_1, \theta_2, \theta_3, \ldots, \theta_i, \ldots \tag{51}$$

Let these have the respective ranges

$$m_0 = m, m_1, m_2, m_3, \ldots, m_i, \ldots \tag{52}$$

and the respective periods

$$r_0 = r, r_1, r_2, r_3, \ldots, r_i, \ldots, \tag{53}$$

where, by definition of derivative,

$$m_{i+1} = r_i. \tag{54}$$

As we shall see in §7, this series of derivatives must terminate after a finite number of steps with endless repetition of the rudimentary substitution (0) on the range 1.

Example: The successive derivatives of $\theta = (0)(2)(4)(6)(1357)$ are $\theta_1 = (0)(2)(13)$, $\theta_2 = (0)(1)$, $\theta_{(3)} = (0)$, $\theta_i = (0)$ for $i > 3$.

We observe the following relations of divisibility:

$$m_{i+2} | m_i, \qquad r_{i+2} | r_i. \tag{55}$$

With use of (54), these follow from the fact that θ_i, θ_{i+1} are conjugate—therefore $r_{i+1} | m_i$; and θ_{i+1}, θ_{i+2} are conjugate—therefore $r_{i+2} | m_{i+1}$.

6. Reduction of a Substitution Modulo a Divisor Range.—Let $\theta(x)$ define a substitution on $[m]$, so that $\theta(x) \equiv \theta(y) \pmod{m}$ is equivalent to $x \equiv y \pmod{m}$. If r is any chosen divisor of m, it may sometimes be the case that $\theta(x) \equiv \theta(y) \pmod{r}$ is equivalent to $x \equiv y \pmod{r}$. When this is so, and only then, $\theta(x)$ determines a substitution $\bar{\theta}$ on the range r, whose representation in cycle notation may be obtained from that of θ by replacing each numeral by its remainder modulo r: $\bar{\theta}(x) \equiv \theta(x) \pmod{r}$. We shall say that θ *reduces to* $\bar{\theta}$ *on the range* r, and write $\theta \rightarrow \bar{\theta} \pmod{r}$.

Examples: By the procedure described, each of the substitutions θ, ϕ in (27) reduces on the range 4 to (0)(2)(0)(2)(1313), i.e., omitting repetition, to (0)(2)(13). The special substitution $\theta = (0)(2)(4)(135)$ on the range 6 reduces on the range 2 to the identity (0)(1). On the other hand, if we attempt to reduce the same substitution modulo 3, we obtain the symbol (0)(2)(1)(102), which does not define a substitution at all—since for this, not only must every given numeral a on the reduced range occur, but every

occurrence must be preceded (cyclically) by the same numeral b and followed (cyclically) by the same numeral c.

If we have a list of special substitutions with their derivatives—that is to say of primary groups—the following theorem enables us to match these substitutions as conjugate pairs. It amounts to a rephrasing of condition (III.1).

THEOREM VI. *Necessary and sufficient conditions for two special substitutions θ, ϕ to be conjugate are:*

(a) the period of each shall divide the range of the other,

(b) each shall reduce modulo the period of the other to the derivative of the other.

In symbols, with our established notation:

(a) $s|m,\ r|n$

(b) $\theta \rightarrow \phi_1 \pmod{s},\ \phi \rightarrow \theta_1 \pmod{r}$.

Since in the series (51), any two consecutive members θ_i, θ_{i+1} are conjugate, it follows that

$$\theta_i \rightarrow \theta_{i+2} \pmod{r_{i+1} = m_{i+2}}. \tag{61}$$

Thus

$$\begin{aligned} &\theta \rightarrow \theta_2 \pmod{m_2},\ \theta_2 \rightarrow \theta_4 \pmod{m_4}, \text{ etc.}, \\ &\theta_1 \rightarrow \theta_3 \pmod{m_3},\ \theta_3 \rightarrow \theta_5 \pmod{m_5}, \text{ etc.} \end{aligned} \tag{62}$$

Accordingly, once the first derivative θ_1 of a special substitution θ has been found by 1-translation of θ^v, the succeeding derivatives can be easily determined by reduction to the appropriate moduli, without further calculation of translations.

7. *Further Properties of Special Substitutions. Type.*—By the *principal cycle* of a substitution ($m > 1$) we shall understand the cycle which contains the numeral 1.

THEOREM VII. *In any special substitution θ on a range $m > 1$, the order of every cycle is a divisor of the order of the principal cycle.*

The following is tantamount:

THEOREM VIII. *For $m > 1$, the period r of any special substitution θ is equal to the order of its principal cycle. A consequence is that*

$$r < m \qquad \text{if} \qquad m > 1. \tag{71}$$

By this and (54), it follows that in the series of derivatives (51) the range continually decreases so long as it stays > 1:

$$m_{i+1} < m_i \qquad \text{if} \qquad m_i > 1. \tag{72}$$

Since we cannot have an infinite descent of integers >1, we must sooner or

later come to a derivative θ_k whose range is $m_k = 1$, i.e., $\theta_k = (0)$ on [1]. If k is the smallest index for which this happens, we shall say that θ is of *type k*. (Of course, θ_i continues $= (0)$ for $i > k$.) In other words: the type k is the order of the lowest derivative which reduces to the rudimentary substitution (0). *Example:* The substitution $\theta = (0)(2)(4)(6)(1357)$ is of type 3 (cf. §5, third paragraph).

Evidently, for $i \leqq k$, the ith derivative θ_i is of type $k - i$.

From (55) and (72) it follows that

$$m_{i+2} \| m_i, \qquad r_{i+2} \| r_i \tag{73}$$

provided $m_i > 1$, $r_i > 1$, respectively. The notation $x \| y$ means that x is an *aliquot* divisor of y: $x | y$ and $x \neq y$.

From (73) we infer

THEOREM IX. *If the range m of a special substitution θ is the product of P prime factors, and the period r the product of Q prime factors, then the type k of θ is subject to the following inequalities:*

$$k \leqq 2P, \; k \leqq 2Q + 1. \tag{74}$$

The following easily proved theorem limits the substitutions which can be paired as conjugates.

THEOREM X. *Conjugate substitutions must be of the same or of consecutive types.*

We conclude this note with a theorem which shows how a pair of conjugate substitutions θ, ϕ can be completely determined when their principal cycles are known.

THEOREM XI. *If the principal cycles of two conjugate substitutions θ, φ are, respectively,* $(a_0, a_1, a_2, \ldots, a_{r-1})$ *and* $(b_0, b_1, b_2, \ldots, b_{s-1})$, *where* $a_0 = 1$, $b_0 = 1$, *then for all* $x \geqq 1$:

$$\theta(x) \equiv a_{b_0} + a_{b_1} + a_{b_2} + \ldots + a_{b_{x-1}} \pmod{m}$$
$$\phi(x) \equiv b_{a_0} + b_{a_1} + b_{a_2} + \ldots + b_{a_{x-1}} \pmod{n}.$$

Illustration: With reference to (27), we find that for $x = 1, 2, 3, \ldots$, the values of $\theta(x)$ are congruent mod 8 to the partial sums of the series $3 + 7 + 3 + 7 + 3 + 7 + \ldots$, and the values of $\phi(x)$ congruent mod 8 to the partial sums of $7 + 3 + 7 + 3 + 7 + 3 + \ldots$.

[1] All numerical variables and functions considered are restricted to *integer* values.

[2] In the main, only 0-fixing substitutions are considered hereafter.

[3] Fixed numerals of a substitution will always be indicated explicitly, so that the range of the substitution may be apparent (1 greater than the maximum numeral).

[4] The vertical line in condition (II) and hereafter denotes "divides."

ON THE BASIS THEOREM FOR FINITE ABELIAN GROUPS (THIRD NOTE)*

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1. Let G denote any finite abelian group; we wish to prove the existence of a basis $(\theta_1, \theta_2, \ldots, \theta_m)$ for G. This is to require (in additive notation) that: (*i*) every element θ of G shall be representable in the form

$$\theta = a_1\theta_1 + a_2\theta_2 + \ldots + a_m\theta_m \tag{1}$$

while (*ii*) (independence condition)

$$c_1\theta_1 + c_2\theta_2 + \ldots + c_m\theta_m = 0 \tag{2}$$

shall imply

$$c_i\theta_i = 0 \qquad \text{for } i = 1, 2, \ldots, m.^1 \tag{3}$$

0 denotes the identical element; small English letters, here and in the sequel, have integer values.

Because of the standard representation of any finite abelian group as direct product of subgroups of prime-power order, the case of prime-power order leads immediately to the general case; one has only to combine the bases of the component subgroups. This justifies us in supposing from now on that the order of G is p^α, p a prime, α a positive integer.

By a B-group we shall understand a subgroup of G that has a basis. Certainly B-groups exist, e.g., the cyclic subgroups of G.

Our goal is to prove the following

THEOREM. *G itself is a B-group.*

We shall use ϵ to denote membership of an element in a group, ϵ non-membership. By $H < K$, or $K > H$, we shall mean that H is a *proper* subgroup of K: H contained in K, but $H \neq K$.

2. LEMMA. *If H is a B-group and $H < G$, then a B-group K exists such that $H < K$.*

Proof: By hypothesis, an element φ of G exists such that $\varphi \epsilon H$. But $p^\lambda\varphi \epsilon H$ if λ is large enough, e.g., if p^λ = period of φ, so that $p^\lambda\varphi = 0$. Let $\lambda > 0$ be fixed so that p^λ is the *lowest* power of p such that $p^\lambda\varphi \epsilon H$. Then

$$p^\lambda\varphi = a_1\theta_1 + a_2\theta_2 + \ldots + a_m\theta_m, \tag{4}$$

where $(\theta_1, \theta_2, \ldots, \theta_m)$ is a basis of H. We suppose this basis to be without zero elements,[2] and written in a descending order (wide sense) of the respective periods:

$$p^{k_1} \geqq p^{k_2} \geqq \ldots \geqq p^{k_m}.$$

Since this is equivalent to

$$k_1 \geqq k_2 \geqq \ldots \geqq k_m,$$

each period is a multiple of all the following ones. A consequence is that $d\theta_r = 0$ implies

$$d\theta_{r+1} = 0,\ d\theta_{r+2} = 0,\ \ldots,\ d\theta_m = 0. \tag{5}$$

In the proof of our lemma there are two cases.

3. *Case I:* Each coefficient a_i in (4) is divisible by p: $a_i = pb_i$ for $i = 1, 2, \ldots, m$.

Then (4) can be written

$$p(p^{\lambda-1}\varphi - b_1\theta_1 - b_2\theta_2 - \ldots - b_m\theta_m) = 0. \tag{6}$$

Let

$$\omega = p^{\lambda-1}\varphi - b_1\theta_1 - b_2\theta_2 - \ldots - b_m\theta_m; \tag{7}$$

then, first, $\omega \in H$; otherwise $p^{\lambda-1}\varphi \in H$, contrary to our least power hypothesis concerning p^λ.

Also

$$p\omega = 0. \tag{8}$$

From this we can infer that $c\omega \in H$ implies $c\omega = 0$. For if $c\omega \neq 0$, p does not divide c; hence $xc + yp = 1$ for certain integers x, y; therefore $\omega = (xc + yp)\omega = x(c\omega) + y(p\omega) = x(c\omega)$; thus $\omega \in H$, contrary to our known condition.

It can now be proved that

$$(\omega, \theta_1, \theta_2, \ldots, \theta_m) \tag{9}$$

are independent elements, and therefore form a basis of the group K which they generate.

For suppose

$$c\omega + c_1\theta_1 + c_2\theta_2 + \ldots + c_m\theta_m = 0.$$

Then $c\omega \in H$, hence (by the second preceding paragraph) $c\omega = 0$; consequently also $c_1\theta_1 = 0, \ldots, c_m\theta_m = 0$, because of the independence of the θ's. Thus all the elements (9) are independent.

Accordingly, K, possessed of a basis, is a B-group. But K contains a basis of H, and therefore H itself—also the element $\omega \in H$. Thus $K > H$.

4. *Case II:* At least one coefficient a_i in (4) is not divisible by p; let a_r be the first such coefficient.

We have then from (4)

$$p(p^{\lambda-1}\varphi - b_1\theta_1 - \ldots - b_{r-1}\theta_{r-1}) = a_r\theta_r + \ldots + a_m\theta_m. \tag{10}$$

Let

$$\omega = p^{\lambda-1}\varphi - b_1\theta_1 - \ldots - b_{r-1}\theta_{r-1}; \tag{11}$$

then $\omega \in H$, otherwise $p^{\lambda-1}\varphi \in H$, contrary to hypothesis (cf. the text just preceding (4)).

Also, by (10),

$$p\omega = a_r\theta_r + a_{r+1}\theta_{r+1} + \ldots + a_m\theta_m, \tag{12}$$

where p does not divide a_r. Therefore integers x, y exist such that

$$xa_r + yp^{k_r} = 1. \tag{13}$$

Here p^{k_r} is the period of θ_r, so that

$$0 = p^{k_r}\theta_r. \tag{14}$$

By linear combination of (12) and (14) with the multipliers x, y, we get

$$xp\omega = \theta_r + xa_{r+1}\theta_{r+1} + \ldots + xa_m\theta_m.$$

It follows that the group K generated by

$$(\theta_1, \theta_2, \ldots, \theta_{r-1}, \omega, \theta_{r+1}, \ldots, \theta_m) \tag{15}$$

includes θ_r, therefore includes all of H.

But K also contains the element $\omega \in H$; hence $K > H$.

It remains to prove that K is a B-group. This we shall do by showing that the generators (15) of K are *independent*, and so form a basis of K.

Indeed, suppose that

$$c_1\theta_1 + c_2\theta_2 + \ldots + c_{r-1}\theta_{r-1} + c\omega + c_{r+1}\theta_{r+1} + \ldots + c_m\theta_m = 0. \tag{16}$$

Then $c\omega \in H$; also $p\omega \in H$, by (12). Hence p divides c: $c = dp$—otherwise $xc + yp = 1$ for certain integers x, y; $\omega = (xc + yp)\omega = x(c\omega) + y(p\omega)$; therefore $\omega \in H$, contrary to the statement following (11).

Consequently, with use of (12),

$$c\omega = dp\omega = da_r\theta_r + da_{r+1}\theta_{r+1} + \ldots + da_m\theta_m. \tag{17}$$

Making this substitution in (16), we find a linear relation in the θ's wherein the term in θ_r is $da_r\theta_r$; by the independence of the θ's, it follows that

$$da_r\theta_r = 0. \tag{18}$$

Since, by (14),

$$dp^{k_r}\theta_r = 0, \tag{19}$$

we obtain with use of (13): $d\theta_r = 0$; hence, by (5),

$$d\theta_{r+1} = 0, \ldots, d\theta_m = 0. \tag{20}$$

By (17), then,

$$c\omega = 0. \tag{21}$$

From the independence of the θ's, it follows that the other terms in (16) are individually $= 0$, thus completing our proof of independence of the elements (15).

5. Let now H_0 be a B-group of *maximum* order; certainly H_0 exists, since the orders of the B-groups are bounded by the finite order of G.

We say $H_0 = G$, thus proving our main theorem.

For if $H_0 < G$, we are in obvious contradiction with our lemma.

* Recent treatments of this "fundamental theorem of abelian groups" are: by the author, these PROCEEDINGS, **37**, 359–362, 525–528 (1951); by Rado, R., *J. London Math. Soc.*, **26** (part I), 74–75 (1951). These new proofs are definitely superior in simplicity and directness to those previously given in the standard treatises on algebra and group theory.

[1] This definition allows any number of zero elements to be adjoined to or removed from a basis. Thus we may always suppose a basis to contain no zero elements—except in the case of the group consisting solely of 0, whose basis we shall regard as 0.

That the elements of a basis (without zero elements) are necessarily distinct follows immediately from the independence condition.

[2] No proof of our lemma is required in the case of the group H consisting solely of 0, for this is properly contained in every other cyclic subgroup of G, i.e., in a B-group.

HARMONIC TENSORS ON MANIFOLDS WITH BOUNDARY

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1. *Introduction.*—The boundary-value problems considered in this note relate to harmonic p-tensors on a Riemannian manifold with boundary. We state certain theorems of existence and uniqueness, which are solutions for the boundary-value problems of Dirichlet and Neumann type for harmonic p-tensors on a boundary manifold. The results which we state, with the exception of Theorem 8, were conjectured by Tucker.[7] Proofs of the theorems will appear in the *Annals of Mathematics*.

We consider alternating (skew-symmetric) covariant tensors $\varphi_{i_1 \ldots i_p}$ of rank p on a Riemannian manifold M with boundary B, and also their associated differential forms,

$$\varphi = \sum_{i_1 < \ldots < i_p} \varphi_{i_1 \ldots i_p} \, dx^{i_1} \wedge \ldots \wedge dx^{i_p}.$$

The differentials dx^i anticommute. The p-forms φ are those considered by Hodge and de Rham in their theory of harmonic integrals on a closed manifold. More recently, de Rham and Kodaira have studied fundamental singularities for harmonic tensors on a manifold, and thus prepared the way for a study of the associated boundary-value problems. The theory of harmonic p-tensors may be considered to be a generalization of the classical potential theory (theory of harmonic functions) in which the first and second boundary-value problems play an important role. Solutions of the corresponding problems for p-tensors are stated in Theorems 6 and 7 of this note. In Section 5 several well-known particular cases of these results are discussed.

2. *Differential Forms.*—Let M be an n-dimensional orientable Riemannian manifold with regular boundary B and positive definite metric. We suppose that the manifold and the metric are C^∞ and that all p-tensors on the manifold are also of the class C^∞. The absolute and relative Betti groups of dimension p, $H_p(M)$, $H_p(M, B)$ associated with M and B are of orders $R_p(M)$ and $R_p(M, B)$, respectively, with $R_p(M, B) = R_{n-p}(M)$.[5] A p-form or p-tensor φ defined on M induces on B a p-tensor $t\varphi$ whose components are precisely those components of φ which are not multiplied by any differential dx^n, where x^n is a local coordinate normal to the boundary B. The p-tensor $t\varphi$ is known as the tangential boundary component of φ, and the residual part of φ, which contains the factor dx^n, is the normal boundary component $n\varphi$. Only on B has this dichotomy an invariant meaning. If $p = 0, t\varphi = \varphi$, if $p = n, t\varphi = 0$.

To each p-tensor φ is associated its differential, a $(p + 1)$-tensor $d\varphi$, with

$$d\varphi = \sum_{i_1 < \ldots < i_p} (d\varphi_{i_1 \ldots i_p})\, dx^{i_1} \wedge \ldots \wedge dx^{i_p}.$$

Let C be a $(p + 1)$-chain of M, and denote its boundary by bC; then the formula of Stokes states that

$$\int_C d\varphi = \int_{bC} \varphi.$$

A form φ for which $d\varphi = 0$ is said to be closed, and a form $\varphi = d\chi$ is said to be derived. Since $d \cdot d = 0$, a derived form is closed. The integral $\int_Z \varphi$ of a closed form φ over a cycle Z of M depends only on the absolute homology class of Z and is known as the period of φ on Z.

The dual $*\varphi$ of a p-tensor φ is an $(n - p)$-tensor defined by

$$(*\varphi)_{j_1 \ldots j_{n-p}} = \sum_{i_1 < \ldots < i_p} e_{i_1 \ldots i_p j_1 \ldots j_{n-p}}\, \varphi_{i_1 \ldots i_p},$$

where

$$e_{i_1 \ldots i_n} = \Gamma_{i_1 \ldots i_n}{}^{12\ldots n} \sqrt{\Gamma_{12\ldots n,\, 12\ldots n}},$$

and

$$\Gamma_{i_1 \ldots i_p,\, j_1 \ldots j_p} = \begin{vmatrix} g_{i_1 j_1} & \cdots & g_{i_1 j_p} \\ \vdots & & \\ g_{i_p j_1} & \cdots & g_{i_p j_p} \end{vmatrix}.$$

For the boundary-value operators there hold the commutation relations $*t = n*$, $*n = t*$. The operation of dual derivation δ is given by

$$\delta\varphi = (-1)^{np+n+1} *d*\varphi.$$

The form $\delta\varphi$ is of degree one less than φ. The Laplace-Beltrami operator Δ for p-vectors is

$$\Delta = d\delta + \delta d.$$

We introduce the scalar product

$$(\alpha, \beta) = \int_M \alpha \wedge *\beta.$$

Since $N(\alpha) = (\alpha, \alpha) > 0$, $\alpha \neq 0$, (α, β) is the scalar product in a real Hilbert space of p-forms on M. From Stokes' formula and the formula for the differential of a product follows the formula of Green for the manifold M:

$$(d\varphi, \psi) - (\varphi, \delta\psi) = \int_B \varphi \wedge *\psi.$$

A p-tensor φ such that $d\varphi = 0$, $\delta\varphi = 0$ is said to be a harmonic field. This definition of a harmonic field coincides with the definition originally used by Hodge. If the less restrictive condition $\Delta\varphi = (d\delta + \delta d)\varphi = 0$ holds, then φ will be termed a Laplace-Beltrami harmonic form, or, more simply, a harmonic form. We also consider p-tensor potentials χ defined by the condition $\delta\, d\chi = 0$. We note that, if χ is a p-tensor potential, then $\varphi = d\chi$ is a harmonic field. If a p-tensor potential χ has the form $\chi = \delta\psi$ then χ is a harmonic form.

3. Closed p-Tensors.—For a closed manifold $M(B = 0)$ it was proved by G. de Rham[6] that there exists a closed form of given degree p with given periods on R_p independent p-cycles of M; and further that a closed form with zero periods is derived. To prove the boundary-value theorems for harmonic fields, we require corresponding results for closed p-forms on manifolds with boundary. These extensions of the de Rham theorems, which we now state, have been proved with the help of de Rham's theorems. For a manifold M with boundary B, we have then

THEOREM 1. *There exists a closed p-tensor with given periods on R_p independent absolute p-cycles.*

THEOREM 2. *There exists a p-tensor whose derivative is a given closed $(p + 1)$-tensor with absolute periods all zero.*

Theorems 1 and 2 are extensions of the de Rham theorems in which conditions are placed on the periods on the absolute cycles; we may call them

"absolute" theorems. For the corresponding relative theorems the following definition is now required. A p-tensor ψ defined on the boundary B is said to be admissible, if

$$d\psi = 0 \qquad \text{on } B,$$
$$\int_R \psi = 0, \qquad R \in H_{p+1}(M, B).$$

THEOREM 3. *There exists a closed p-tensor having a given admissible tangential boundary value, and given periods on R_{n-p} given independent relative p-cycles.*

THEOREM 4. *There exists a p-tensor whose tangential boundary value is a given p-tensor ψ and whose derivative is a given exact (closed) $(p+1)$-tensor χ such that*

$$t\chi = d\psi$$
$$\int_R \chi = \int_{bR} \psi, \qquad R \in H_{p+1}(M, B).$$

It is clear from Stokes' formula that a p-tensor φ closed in M has a tangential boundary value $t\varphi$ which is admissible. Theorem 3 states that, conversely, an admissible tangential boundary value can be extended to a p-form closed in M. Theorems 3 and 4 give relative extensions of both de Rham theorems. The differentiable structure of the manifold is sufficient to ensure the validity of the results, no metric being required.

4. The Existence of p-Tensor Potentials with Given Boundary Values.—Under the conditions laid down in paragraph 2, a result, which may be stated as follows, holds for the manifold M with boundary B:

THEOREM 5. *Let ψ be a given p-tensor, norm-finite and C^∞ on M and B but otherwise arbitrary. Then there exists a p-tensor potential φ ($\delta d\varphi = 0$) on M such that $t\varphi = t\psi$ on B. If also $\psi = \delta\chi$, there exists a unique p-tensor potential $\varphi = \delta\zeta$ satisfying $t\varphi = t\psi$ on B.*

The proof of Theorem 5 is based on the fact that every finite submanifold of an orientable Riemannian manifold may be imbedded in a closed orientable Riemannian manifold. On the closed manifold we have the de Rham kernel $g_p(x, y)$ satisfying $\Delta_x g_p(x, y) \equiv -\alpha_p(x, y)$, where $\alpha_p(x, y)$ is the reproducing kernel for harmonic fields on the closed manifold, and $\delta_x g_p(x, y)$ is a fundamental singularity for the operator δd on the submanifold. With this fundamental singularity, the method of integral equations developed in reference 1 for Euclidean space may be applied to establish Theorem 5.

Dual to Theorem 5 is a similar result for dual p-tensor potentials ψ satisfying the condition $d\delta\psi = 0$. A result similar to Theorem 5 was established by Hodge for the case when M is a cell in a flat space.[2a] The present theorem is then an extension of Hodge's result to a general C^∞ Riemannian manifold.

5. The Existence and Uniqueness Theorems for Harmonic Fields.—We state first the result which constitutes a solution for the Dirichlet problem for harmonic fields on M.

THEOREM 6. *There exists a unique harmonic field φ having a given admissible tangential boundary value and given periods on $R_p(M, B)$ given independent relative p-cycles of M.*

Dual to this result is the solution for the Neumann problem, expressed by

THEOREM 7. *There exists a unique harmonic field φ whose dual has a given admissible tangential boundary value, and which itself has given periods on $R_p(M)$ independent absolute p-cycles of M.*

The proofs of Theorems 6 and 7 make use of Theorems 1 through 5. Omitting the proofs, we mention here some special cases of the results. If the manifold M is closed, both theorems reduce to the well-known result of Hodge, namely, that there exists a unique harmonic field with given periods on R_p independent p-cycles.[2b] If $n = 2$, in which case the Riemannian manifold becomes locally conformally flat, we may deduce from the case $p = 1$ existence theorems for analytic differentials on a Riemann surface, and for the Dirichlet and Neumann problems for harmonic functions Many years ago Kelvin stated a result which is the case $n = 3$, $p = 1$ of Theorem 7 in Euclidean space, and which is the first such result to show how the topological properties of the space, as well as the boundary values, appear in the result. Kelvin's theorem states that the irrotational motion of an incompressible fluid in a multiply connected region of 3-dimensional space is determined when the normal velocity component at the boundary and the circulations in the independent circuits of the region are given.[3]

6. Harmonic Forms.—We conclude with some remarks on forms φ satisfying $\Delta\varphi = 0$, $(\Delta = d\delta + \delta d)$. If M is a closed manifold, and $\Delta\varphi = 0$, then $d\varphi = 0$ and $\delta\varphi = 0$, as can be verified from Green's formula. In a manifold with boundary this result is not true in general. Kodaira[4] and de Rham have established the existence of a local fundamental singularity for the equation $\Delta\varphi = 0$. However, it is not known that a fundamental singularity in the large exists unless the conditions $\Delta\varphi = 0, t\varphi = 0, n\varphi = 0$ imply $\varphi \equiv 0$ in M. These conditions do imply, however, that $d\varphi = 0$ and $\delta\varphi = 0$; so that in view of Theorems 6 and 7, the condition is satisfied if, for instance, either the relative or absolute homology group of dimension p of M is trivial.

Assuming that the condition is satisfied, we have

THEOREM 8. *There exists a unique harmonic form φ on M with given tangential and normal boundary values.*

The result is an extension of a classical theorem for harmonic functions, and the proof follows well-known lines.

[1] Garabedian, P. R., and Spencer, D. C., "Complex Boundary Value Problems," Technical Report No. 16, Stanford University, 1951.

[2] Hodge, W. V. D., (*a*) *J. Lond. Math. Soc.*, **36** (2), 257 (1933). (*b*) *Theory of Harmonic Integrals*, Cambridge, 1941.

[3] Lord Kelvin, *Papers*, **4**, 13, Cambridge University Press.

[4] Kodaira, *Ann. Math.*, **50**, 587 (1949).

[5] Lefschetz, S., *Topology*, New York, 1930.

[6] de Rham, G., *J. Math.*, 115–200 (1931).

[7] Tucker, A. W., *Bull. Am. Math. Soc.*, **47**, 714 (1941).

A SUMMARY OF NEW RESULTS CONCERNING THE SOLUTIONS OF EQUATIONS IN FINITE FIELDS

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Communicated by H. S. Vandiver, July 9, 1951

In this paper we shall give a summary of several results we have obtained concerning the number of distinct sets of solutions of equations of the type

$$c_1 x_1^{m_1} + c_2 x_2^{m_2} + \ldots + c_s x_s^{m_s} + c_{s+1} = 0, \tag{1}$$

$s \geqq 2$, where the c's are given elements of a finite field of order p^n, p an odd prime, which will be designated $F(p^n)$ and $p^n - 1 = q_i m_i$, $i = 1, \ldots, s$, with $c_1 \ldots c_s \neq 0$ in $F(p^n)$. In the case $c_{s+1} \neq 0$ we shall consider the equation

$$g^{y_1 m_1 + r_1} + g^{y_2 m_2 + r_2} + \ldots + g^{y_s m_s + r_s} = 1 \tag{2}$$

where g is a generator of $F(p^n)$ aside from zero and $-c_i/c_{s+1} = g^{h_i m_i + r_i}$. Let $(r_1, \ldots, r_s)$ denote the number of distinct sets $y_1, \ldots, y_s$; $y_i = 0, 1, \ldots, q_i - 1$, that satisfy equation (2); thus $(r_1, \ldots, r_s) m_1 \ldots m_s$ is the number of distinct sets of non-zero solutions of (1).

H. S. Vandiver[1] gave an expression for the number of solutions of (2),

$$(r_1, \ldots, r_s) \prod_{i=1}^{s} m_i = \sum_{u_1, \ldots, u_s} \psi(\alpha_1^{u_1}, \ldots, \alpha_s^{u_s}) \prod_{i=1}^{s} \alpha_i^{-u_i r_i} \tag{3}$$

where u_i ranges from 0 to $m_i - 1$, $i = 1, \ldots, s$. For $s > 1$,

$$\psi(\alpha_1^{u_1}, \ldots, \alpha_s^{u_s}) = \sum_{a_1, \ldots, a_{s-1}} \alpha_s^{u_s \operatorname{ind} A} \prod_{i=1}^{s-1} \alpha_i^{u_i \operatorname{ind} a_i}, \tag{4}$$

$$\psi(\alpha_i^{u_i}) = 1 \tag{4a}$$

where $\alpha_i = e^{2\pi i/m_i}$; ind a_i is defined $g^{\operatorname{ind} a_i} = a_i$ in $F(p^n)$ with g a generator of the cyclic group formed by the non-zero elements of $F(p^n)$ under multiplication; $\alpha_i^{u_i \operatorname{ind} 0} = 0$ for all u_i; and each a ranges independently over each non-zero element of $F(p^n)$ with $A = 1 - a_1 - \ldots - a_{s-1}$.

By reducing the degenerate cases of (4), cases where $|\psi| \neq p^{(s-1)n/2}$, (see reference 2), we obtain

THEOREM I.

$$(r_1, \ldots, r_s) \prod_{i=1}^{s} m_i = p^{(s-1)n} - \sum_{t=1}^{s-1} S_t^s (r_1, \ldots, r_t) \prod_{i=1}^{t} m_i + B(r_1, \ldots, r_s) \quad (5)$$

where

$$B(r_1, \ldots, r_s) = \sum_{u_1, \ldots, u_s} \psi(\alpha_1^{u_1}, \ldots, \alpha_s^{u_s}) \prod_{i=1}^{s} \alpha_i^{-u_i r_i} \quad (6)$$

with u_i ranging over the set $1, 2, \ldots, m_i - 1$, and

$$S_t^s F(b_1, \ldots, b_t)$$

denotes the sum of the F's of the $\binom{s}{t}$ distinct sets of b's taken t at a time.

If we denote the number of solutions of (1) by

$$[r_1, \ldots, r_s], \quad (7)$$

we have from Theorem I,

COROLLARY.

$$[r_1, \ldots, r_s] = p^{(s-1)n} + B(r_1, \ldots, r_s) \quad (8)$$

where $B(r_1, \ldots, r_1)$ is defined in (6).

From (8) we obtain the following limits, (see reference 3),

$$\left|[r_1, \ldots, r_s] - p^{(s-1)n}\right| \leqq p^{(s-1)n/2} \left(\prod_{i=1}^{s} (m_i - 1) - K \right) + p^{(s-2)n/2} K \quad (9)$$

where K is the number of sets of u's in the sets $1, \ldots, m_i - 1$, $i = 1, \ldots, s$, such that $\alpha_1^{u_1} \ldots \alpha_s^{u_s} = 1$.

If M is the least common multiple of $m_1, \ldots, m_s$, we obtain from relation (3)

$$(r_1, \ldots, r_s) \prod_{i=1}^{s} m_i \leqq ((p^n - 1)^s - 1 + (-1)^{s+1})/p^n + ((p^{n/2}(M-1) + 1)^{s+1} + (M-1)(p^{n/2} + 1)^{s+1})/Mp^n. \quad (10)$$

From Corollary I

$$\sum_{r_i=0}^{m_i - 1} [r_1, \ldots, r_s] = p^{(s-1)n} m_i \quad (11)$$

follows easily.

Also from Corollary I we obtain, (see reference 4),

THEOREM II.

$$\sum_{r_2, \ldots, r_s} [r_1, \ldots, r_s][r_1 + h_1, \ldots, r_s + h_s] = p^{2(s-1)n} \prod_{i=1}^{s} m_i +$$

$$\begin{cases} p^{(s-1)n} \prod_{i=1}^{s} m_i \prod_{i=1}^{t} (m_i - 1)(-1)^{s-t}, & \begin{cases} h_i \equiv 0 \pmod{m_i}, & i \leq t \\ h_i \not\equiv 0 \pmod{m_i}, & i > t \end{cases} \\ p^{(s-1)n} \prod_{i=1}^{s} m_i(-1)^s, \; h_i \not\equiv 0 \pmod{m_i}, \; i = 1, \ldots, s \end{cases} +$$

$$(p^{(s-2)n} - p^{(s-1)n}) \prod_{i=1}^{s} m_i \sum_{u_1, \ldots, u_s} \alpha_1^{-u_1 h_1} \ldots \alpha_s^{-u_s h_s}$$

where the u's range over the sets 1, . . ., $m_i - 1$, $i = 1, \ldots, s$, such that $\alpha_1^{u_1} \ldots \alpha_s^{u_s} = 1$; and the r's range over the sets 0, 1, . . ., $m_i - 1$, $i = 1, \ldots, s$.

We obtain by direct addition from (3)

THEOREM III.

$$\sum_{r_{s-t+1}, \ldots, r_s} (r_1, \ldots, r_s) \prod_{i=1}^{s-t} m_i = (p^n - 1)^{s-t}((p^n - 1)^t - (-1)^t)/p^n +$$

$$(-1)^t (r_1, \ldots, r_{s-t}) \prod_{i=1}^{s-t} m_i \quad (12)$$

where $s - t + 1 > 2$ and r_i range over the sets 0, 1, . . ., $m_i - 1$, $i = s - t + 1$, . . ., s. (13)

COROLLARY.

$$\sum_{r_2, \ldots, r_s} (r_1, \ldots, r_s) m_1 = (p^n - 1)((p^n - 1)^{s-1} - (-1)^{s-1})/p^n +$$

$$(-1)^{s-1} \begin{cases} 0, & r_1 \not\equiv 0 \pmod{m_1} \\ m_1, & r_1 \equiv 0 \pmod{m_1} \end{cases}$$

where r_i range over the sets 0, 1, . . ., $m_i - 1$, $i = 2, \ldots, s$.

If $m_1 = m_2 = \ldots = m_s$, n even and $p^{n/2} + 1 \equiv 0 \pmod{m_1}$, then we have obtained the exact values of $(0, \ldots, 0)m_1^s$ and $[0, \ldots, 0]$ in terms of s, p, n and m_1 explicitly.

If the m's in (1) are grouped into k sets which are prime each to each with $c_{s+1} = 0$, we have an equation of the form

$$\sum_{i=1}^{s_1} c_{1,i} x_{1,i} u^{m_{1,i}} + \sum_{i=1}^{s_2} c_{2,i} x_{2,i}^{m_{2,i}} + \ldots + \sum_{i=1}^{s_k} c_{k,1} x_{k,i}^{m_{k,i}} = 0 \quad (14)$$

Let m_t be the least common multiple of $m_{t,i}$ for each t and $i = 1, \ldots, s_t$; thus $(m_h, m_j) = 1$, $h \neq j$. Also impose the condition that $m_t = m_{t,1}$, $t < k$. Let H_t denote the number of distinct non-zero solutions of

$$\sum_{i=1}^{s_t} c_{t,i} x_{t,i}^{m_{t,i}} = 0.$$

THEOREM IV. *If N_k denotes the number of distinct sets of non-zero solutions in $F(p^n)$ of (14) then*

$$N_k = (1/p^n)(p^n - 1)^{s_1 + \ldots + s_k} + (-1)^k \prod_{i=1}^{k} ((p^n - 1)^{s_i} - p^n H_i)/p^n(p^n - 1)^{k-1}.$$

From Theorem IV we may in a great variety of cases find the exact values of N_k; such as trinomial equations of type (1) with $c_{s+1} = 0$ and one exponent prime to the other two.

[1] Vandiver, H. S., "On a Generalization of a Jacobi Exponential Sum Associated with Cyclotomy," these PROCEEDINGS, **36,** 144–151 (1950).

[2] Faircloth, O. B., and Vandiver, H. S., "On Multiplicative Properties of a Generalized Jacobi-Cauchy Cyclotomic Sum," *Ibid.*, **36,** 260–267 (1950).

[3] Weil, A., "Numbers of Solutions of Equations in Finite Fields," *Bull. Am. Math. Soc.*, **55,** 497–508 (1949). This limit is an improvement over that given by Weil, p. 502.

[4] Whiteman, A. L., "Finite Fourier Series and Cyclotomy," these PROCEEDINGS, **37,** 373–378 (1951). In this paper Whiteman gives analogous quadratic relations involving the parentheses symbols.

COMPACT SUBGROUPS

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THEOREM: *Every connected compact subgroup of a locally compact group is contained in a maximal connected compact subgroup.*

For connected (L)-groups (locally compact groups which are projective limits of Lie groups) Iwasawa[1] has shown the existence of maximal compact subgroups and that all of these are conjugate. His proof relies on analytic investigation of the approximating Lie groups. Our proof uses only methods of group theory and topology and leads to a much weaker result, which, however, strengthens the standing conjecture that all connected locally compact groups are (L)-groups.

LEMMA 1. *Let $\{H_\alpha\}$ be an increasing collection of connected closed subgroups of a locally compact group G. Let U be a neighborhood of e in G. Then there exists an index β such that $H_\alpha \subset H_\beta U$ for all α.*

Proof: Without loss of generality we assume U compact. If the conclusion is false, then we can choose from $\{H_\alpha\}$ an infinite sequence (which we reindex) $H_0 \subset H_1$ non-$\subset H_2 \subset \ldots$ such that H_i non-$\subset H_{i-1}U$ for $i = 1, 2, \ldots$ Assuming such a sequence we shall deduce a contradiction.

Let V be another neighborhood of e such that $V^2 \subset U$. For each $i = 1, 2, \ldots$ we shall find an element h_i such that $h_i \in U \cap H_i$ and h_i non-$\in H_{i-1}V$. Let φ be the natural mapping of H_i onto the right coset space H_i/H_{i-1}. Now $\varphi(U \cap H_i)$ does not cover H_i/H_{i-1}, for if it did we should have

$H_i \subset \varphi^{-1}\varphi(U \cap H_i) = H_{i-1}(U \cap H_i) \subset H_{i-1}U$. H_i/H_{i-1} is connected, so we can choose h_i so that $\varphi(h_i)$ is a boundary point of $\varphi(U \cap H_i)$. If $h_i \epsilon H_{i-1}V$ then $h_iV \subset H_{i-1}U$ and, relative to H_i, h_i would be an interior point of the sets $h_iV \cap H_i \subset H_{i-1}U \cap H_i = H_{i-1}(U \cap H_i)$; whence, φ being an open mapping, $\varphi(h_i)$ would be interior to $\varphi(H_{i-1}(U \cap H_i)) = \varphi(U \cap H_i)$. Thus, we have obtained an infinite sequence of elements of U which satisfy the relation $h_i^{-1}h_j$ non-ϵ V for $i < j$. This contradicts the compactness of U.

LEMMA 2. *If $\{H_\alpha\}$ is an increasing collection of connected compact subgroups of a locally compact group G, then $(\cup H_\alpha)^-$ is a connected compact subgroup.*

Proof: That it is a connected subgroup is trivial. Choose a compact neighborhood U of e in G, and an index β as in Lemma 1. Then $(\cup H_\alpha)^-$ is compact being a closed subset of $H_\beta U$.

The theorem now follows directly from Zorn's Lemma. In the proof we have used only the connectedness of certain factor spaces; hence we can strengthen our result slightly:

COROLLARY. *Every compact subgroup H of a locally compact group is contained in a compact subgroup which is maximal among those compact subgroups K for which K/H is connected.*

[1] Iwasawa, K., "On Some Types of Topological Groups," *Ann. Math.*, **50**, 507–558 (1949).

DIFFERENTIAL INVARIANTS AND INVARIANT PARTIAL DIFFERENTIAL EQUATIONS UNDER CONTINUOUS TRANSFORMATION GROUPS IN NORMED LINEAR SPACES

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1. Introduction.—The object of this paper is to announce some fundamental results on a general invariant theory of certain continuous transformation groups in normed linear spaces. The main results are novel even if the normed linear spaces are finite dimensional. The comparatively "simplest case" of one-dimensional normed linear spaces and continuous groups in only one numerical parameter *envisages remarkable applications to the solution of problems in aeronautics and related fields.* We shall, however, in no way go into these applications in this paper. For *some* of these possibilities we can refer the reader to a 1951 Ph.D. thesis by A. J. A. Morgan of the Guggenheim Graduate School of Aeronautics at C.I.T. The thesis was written under my direction and with my collaboration.

2. The Continuous Transformation Group.—Let G_r be the r-parameter continuous transformation group

$$\left.\begin{aligned} \bar{x}^i &= f^i(x^1, \ldots, x^m, a^1, \ldots, a^r) \\ \bar{y} &= \phi(y, a^1, \ldots, a^r) \\ (i &= 1, 2, \ldots, m \text{ with } m \geq 2, r \geq 1). \end{aligned}\right\} \tag{2.1}$$

For the purposes of the present exposition, we shall restrict ourselves to transformation groups G_r of Class C_n (existence and continuity of the nth order total Fréchet differentials in open sets with n sufficiently large) with the x's and a's numerical and the y *a variable in a Banach space* B. We shall use the notations $\bar{x} = f(x, a)$, $\bar{y} = \phi(y, a)$ and other similar abbreviations to shorten the exposition.

We shall *assume that the* r*-parameter subgroup of transformations* $\bar{x} = f(x, a)$ *possesses* $s \geq 1$ *functionally* independent Fréchet *differentiable scalar invariants* $\eta_1(x^1, \ldots, x^m), \ldots, \eta_s(x^1, \ldots, x^m)$. Clearly $s < m$. Finally we shall *restrict ourselves to transformation groups* G_r *that possess a B-valued Fréchet differentiable scalar invariant* $g(y, x^1, \ldots, x^m)$ *solvable in* y *for all admissible* y *and* x's *in the group.*

It is convenient to make the following

DEFINITION. *A B-valued function* $y(x^1, \ldots, x^m)$ *of the numerical variables* $x^1, \ldots, x^m$ *will be called an "invariant function under* G_r*" if*

$$y(\bar{x}) = \phi(y(x), a),$$

i.e., the transform of the function $y(x)$ *under* G_r *has the same form as a function of the transformed variables* $\bar{x}^1, \ldots, \bar{x}^m$.

3. Two Fundamental Theorems on Differential Invariants of G_r.—We shall now state and outline briefly the proof of the following fundamental.

THEOREM 1. *If* B_1 *and* B *are two reflexive Banach spaces (not necessarily distinct) and if*

$$\Omega\left(x, y(x), \frac{\partial y}{\partial x}, \ldots, \frac{\partial^q y}{\partial x^q}\right) \tag{3.1}$$

is a B_1*-valued* q*th order differential invariant of invariant B-valued functions* $y(x^1, \ldots, x^m)$ *under the group* G_r, *then* Ω *is expressible in the form*

$$\omega\left(\eta, \nu(\eta), \frac{\partial \nu}{\partial \eta}, \ldots, \frac{\partial^q \nu}{\partial \eta^q}\right), \tag{3.2}$$

where the corresponding B-valued functions $\nu(\eta_1, \ldots, \eta_s)$ *exist and are determined by the formula*

$$\nu(\eta_1(x^1, \ldots, x^m), \ldots, \eta_s(x^1, \ldots, x^m)) = g(y(x^1, \ldots, x^m), x^1, \ldots, x^m). \tag{3.3}$$

Outline of Proof. If $y(x)$ is an invariant B-valued function under the group G_r, then by the reflexivity of the Banach space B, one can show that there exists a B-valued function $\nu(\lambda_1, \ldots, \lambda_s)$ of s numerical variables $\lambda_1, \ldots, \lambda_s$ such that the identity (3.3) holds. Now there exists a set of m auxiliary variables ${}^*x^i$ $(i = 1, 2, \ldots, m)$ in terms of which $x^1, \ldots, x^m$ can be expressed such that

$$y(x^1, \ldots, x^m) = \gamma({}^*x^{s+1}, \ldots, {}^*x^m, \eta, \nu(\eta)) \tag{3.4}$$

for some B-valued function γ. Moreover, one can show on using (3.4) (3.3) that

$$\frac{\partial \nu(\eta)}{\partial \eta_j} \frac{\partial \eta_j}{\partial {}^*x^i} = g\left(y(x), x; \frac{\partial y}{\partial {}^*x^i}\right) + \left(\frac{\partial g(y(x), x)}{\partial {}^*x^i}\right)_{y, x} \tag{3.5}$$

$(i = 1, 2, \ldots, s).$

Then from the solvability of the partial Fréchet differential $g(y, x; \delta y)$ of $g(y, x)$ in y considered as a solvable linear function of the increment δy, we have

$$\frac{\partial y(x)}{\partial {}^*x^i} = \psi_i\left({}^*x^{s+1}, \ldots, {}^*x^m, \eta, \nu(\eta), \frac{\partial \nu}{\partial \eta}\right) \qquad (i = 1, 2, \ldots, s) \tag{3.6}$$

for some s B-valued functions ψ_i. On using (3.5) and well-known theorems on Fréchet differentials we obtain

$$\left.\begin{aligned} &\frac{\partial^l \nu(\eta)}{\partial \eta_{j_1} \ldots \partial \eta_{j_l}} \frac{\partial \eta_{j_1}}{\partial {}^*x^{i_1}} \cdots \frac{\partial \eta_{j_l}}{\partial {}^*x^{i_l}} + (*) = \\ &g\left(y(x), x; \frac{\partial^l y}{\partial {}^*x^{i_1} \ldots \partial {}^*x^{i_l}}\right) + (**) \qquad (i_1, \ldots, i_l = 1, \ldots, s), \end{aligned}\right\} \tag{3.7}$$

where the terms in $(*)$ on the left contain lower derivatives of ν than the lth while the terms in $(**)$ on the right contain y, derivatives of y of lower order than l, and Fréchet differentials of $g(y, x)$ of higher order than one. Consequently,

$$\frac{\partial^l y(x)}{\partial {}^*x^{i_1} \ldots \partial {}^*x^{i_l}} = \psi_{i_1 \ldots i_l}\left({}^*x^{s+1}, \ldots, {}^*x^m, \eta, \nu(\eta), \frac{\partial \nu}{\partial \eta}, \ldots, \frac{\partial^l \nu}{\partial \eta^l}\right)$$

$$(i_1, \ldots, i_l = 1, \ldots, s \text{ and } l \geq 1) \tag{3.8}$$

for some B-valued functions $\psi_{i_1 \ldots i_l}$.

With the aid of (3.4), (3.8) and the reflexivity of the Banach space B_1, it can be shown that (3.1) can be expressed in the form (3.2).

The following partial converse to Theorem 1 can be established. It is to be observed that the reflexivity of the Banach spaces is not needed here.

THEOREM 2. *If B_1 and B are two Banach spaces (not necessarily distinct) and if $v(\eta_1, \ldots, \eta_s)$ is an arbitrary B-valued function of the s invariants $\eta_1, \ldots, \eta_s$, then the corresponding B-valued function $y(x^1, \ldots, x^m)$ determined by formula (3.3) is an invariant function under G_r, and moreover an arbitrary B_1-valued qth order differential expression (3.2) is expressible as a B_1-valued qth order differential invariant (3.1) of invariant functions $y(x^1, \ldots, x^m)$ under the continuous group G_r.*

4. Invariant Partial Differential Equations.—The following important theorem is a consequence of the preceding two theorems. The quest of invariant solutions $y(x^1, \ldots, x^m)$ of certain partial differential equations (4.1) in normed linear spaces is thus reduced to the problem of the solution of a partial differential equation (4.2) of the same order q in m-s smaller number of independent variables. Clearly, if $m = 2$, then $s = 1$ and (4.2) is a qth order "ordinary" differential equation in normed linear spaces.

THEOREM 3. *Under the hypotheses of Theorem 1, an invariant solution $y(x^1, \ldots, x^m)$ (i.e., a solution that is an invariant function under the group G_r) of the partial differential equation.*

$$\Omega\left(x,\ y(x),\ \frac{\partial y}{\partial x},\ \ldots,\ \frac{\partial^q y}{\partial x^q}\right) = 0 \tag{4.1}$$

determines by means of (3.3) a solution $v(\eta_1, \ldots, \eta_s)$ of the corresponding partial differential equation

$$\omega\left(\eta,\ v(\eta),\ \frac{\partial v}{\partial \eta},\ \ldots,\ \frac{\partial^q v}{\partial \eta^q}\right) = 0. \tag{4.2}$$

Conversely, to each solution $v(\eta_1, \ldots, \eta_s)$ of the same (4.2) there corresponds an invariant solution $y(x^1, \ldots, x^m)$ of (4.1).

5. Some Further Topics and Generalizations.—The above theorems are indicative of some results in what is a large subject. Anyone acquainted with the author's general differential geometries with abstract coordinates (see, for example, *Bull. Am. Math. Soc.*, 1939) can "geometrize" the subject by tying it down to topological spaces with abstract normed linear *coordinates*.

The theory of analytic functions of variables in complex Banach spaces begun by the author and R. S. Martin in 1931 has been studied and used by many authors in many remarkable investigations in more recent years. If the Banach spaces in the theory of the group G_r are taken to be complex Banach spaces and if the other numerical variables and parameters are complex, then we clearly have abstract analytic transformation groups G_r. There thus results another important application of the theory of analytic functions of abstract variables.

There are several generalizations both in the type of transformation

group G_r and in the character of the differential invariants and differential equations. We shall make a few remarks on one such generalization. Let $E_1, \ldots, E_f$ be f Banach spaces and $B_1, \ldots, B_g$ be also g Banach spaces. In the place of the group G_r we shall take the continuous r-parameter transformation group Γ_r:

$$\left.\begin{aligned} \bar{x}^i &= f^i(x^1, \ldots, x^m, a^1, \ldots, a^r) \\ \bar{y}_\alpha &= \phi_\alpha(y_\alpha, a^1, \ldots, a^r) \\ (i &= 1, 2, \ldots, m \text{ with } m \geq 2,\ r \geq 1 \text{ and } \alpha = 1, 2, \ldots, g), \end{aligned}\right\} \tag{5.1}$$

where $y_\alpha \in B_\alpha$. Furthermore, in generalizing the differential invariants and invariant differential equations, we can consider f *relative* qth order differential invariants, i.e., f functions Ω_λ of the x's, y's and the derivatives of the y's up to and including the q_λth with the property of retaining their form under the group Γ_r within a corresponding numerically valued factor function ρ_λ. More specifically, we have under Γ_r

$$\begin{aligned} \Omega_\lambda\left(\bar{x}, \bar{y}(\bar{x}), \ldots, \bar{y}_g(\bar{x}), \frac{\partial \bar{y}_1}{\partial \bar{x}^1}, \ldots, \frac{\partial^{q_\lambda} \bar{y}_g}{\partial \bar{x}^{m q_\lambda}}\right) = \\ \rho^\lambda\left(x, y_1, \ldots, y_g, \frac{\partial y_1}{\partial x^1}, \ldots, \frac{\partial^{q_\lambda} y_g}{\partial x^{m q_\lambda}}, a^1, \ldots, a^r\right) \Omega_\lambda \\ \left(x, y_1(x), \ldots, y_g(x), \frac{\partial y_1}{\partial x^1}, \ldots, \frac{\partial^{q_\lambda} y_g}{\partial x^{m q_\lambda}}\right) \end{aligned} \tag{5.2}$$

$$(\lambda = 1, 2, \ldots, f).$$

Of course, there are other types of relative differential invariants. However, the essential generalization in the above type, results from the possibility of allowing f distinct factors ρ_λ and f distinct orders q_λ in (5.2). For the generalized groups Γ_λ and their relative differential invariants Ω_λ, one can prove, among others, theorems corresponding to Theorem 1, Theorem 2 and Theorem 3.

A FACTORIAL STUDY OF THE SUPREME COURT

By L. L. Thurstone and J. W. Degan

Psychometric Laboratory, University of Chicago

Read before the Academy, April 23, 1951

The object of the present study is to ascertain by factorial methods whether the voting records of the individual judges give evidence of any groupings in the Supreme Court. If these can be found, it should be possible to make prediction of the votes of individual judges on some particular issues. Ideally it might be supposed that the judges make completely impartial and impersonal legal judgments about legislation. No group of human beings can ever be expected to perform that way. It can be taken for granted that, no matter how the judges are selected, there will be differences among them as to their loyalties, their political and social philosophies and their attitudes on economic issues. It can be taken for granted that these differences in attitudes and points of view will have an effect on their individual decisions.

There is increasing interest in the application of scientific method to the study of social phenomena. One of the requirements of a scientific method is that it shall be relatively free from the value judgments of the investigator. The interpretation of scientific work by any method is always essentially subjective. The gain with scientific method is that it restricts the range for subjective interpretation to that which is not denied by empirical demonstration. No scientific hypothesis can ever be proved to be correct. At best, it can only be shown to be plausible. It is our belief that social phenomena can eventually be studied with the rigor that characterizes the older sciences, and that social phenomena are not necessarily more complex than physical phenomena. This paper is an exploration in scientific method on the problem of identifying the blocks or subgroups within a larger group as in a legislature, council, or court, in terms of the voting records of the members.

The present study was made on the voting records of the judges during the 1943–1944 and 1944–1945 terms of the Court on 115 cases in which there were at least two dissenting votes. The cases in which the Court was unanimous evidently could not give any indication of differences among the judges and hence those cases were eliminated. A correlation coefficient was computed for each pair of judges. With nine judges there were thirty-six such correlations and these are shown in table 1. A positive correlation coefficient shows that the two judges tended to agree in their votes, whereas a negative coefficient shows that the judges tended to disagree. These coefficients are of some interest. A fourfold table was prepared for each pair of judges. The majority judgment of the court was

taken as a reference, so that the two categories of classification for each vote were: (1) agreement with the majority of the Court, and (2) dissent from the majority of the court. The coefficients represent intraclass phi-coefficients.

The correlation matrix was factored by the complete centroid method. The factoring was repeated three times to stabilize the communalities.

TABLE 1

INTRACLASS ϕ COEFFICIENTS R

		1	2	3	4	5	6	7	8	9
Black	1	..	0.59	0.42	0.40	−0.15	−0.25	−0.41	−0.46	−0.66
Douglas	2	0.59	..	0.30	0.35	−0.10	−0.16	−0.36	−0.45	−0.60
Rutledge	3	0.42	0.30	..	0.46	−0.19	−0.21	−0.36	−0.38	−0.50
Murphy	4	0.40	0.35	0.46	..	−0.25	−0.28	−0.42	−0.32	−0.39
Reed	5	−0.15	−0.10	−0.19	−0.25	..	−0.13	0.29	0.07	−0.07
Jackson	6	−0.25	−0.16	−0.21	−0.28	−0.13	..	−0.05	0.26	0.03
Stone	7	−0.41	−0.36	−0.36	−0.42	0.29	−0.05	..	0.19	0.23
Frankfurter	8	−0.46	−0.45	−0.38	−0.32	0.07	0.26	0.19	..	0.40
Roberts	9	−0.66	−0.60	−0.50	−0.39	−0.07	0.03	0.23	0.40	..

The final orthogonal factor matrix is shown in table 2. Here the common factor variance of the nine variables in the initial table has been represented in three common factors, which are represented by the first three columns of table 2. Table 3 shows the residual variance which is quite small. We conclude, therefore, that the common factor variance in the initial correlation matrix can be accounted for fairly well by three common factors.

TABLE 2

CENTROID FACTOR MATRIX F

	I	II	III	h^2
1	0.80	0.23	0.24	0.75
2	0.70	0.29	0.31	0.67
3	0.64	−0.10	−0.10	0.43
4	0.67	−0.24	−0.17	0.54
5	−0.21	0.45	−0.06	0.25
6	−0.28	−0.23	0.58	0.47
7	−0.53	0.38	−0.23	0.48
8	−0.57	−0.19	0.08	0.37
9	−0.64	−0.23	−0.10	0.47

The total common factor variance is represented in the last column, h^2, of table 2. At this stage of the analysis some interesting information is available. If there were no groupings among the judges to influence their votes, then the rank of the matrix in table 1 would be fairly high. It would not be so low as 3. The last column of table 2 shows the communality for each judge. The two highest values are for Judges Black and Douglas, which indicate that they have high consistency in their voting. The

lowest values in the fourth column are for Judges Reed and Frankfurter, whose votes should be expected to be less consistent with respect to the common factors in the voting.

It is common practice to give a geometrical interpretation of the factor matrix. In most factorial studies the number of common factors is greater than three, so that geometrical models cannot be made. In the present case, where the dimensionality is only three, it is possible to make a geometrical model of the factorial structure in table 2. In the present case the model would be three-dimensional and it would consist of nine vectors, one for each of the judges. Each vector would be defined by three coordinate numbers as given in table 2. The length of each vector would be the square root of the communality which is shown in the last column.

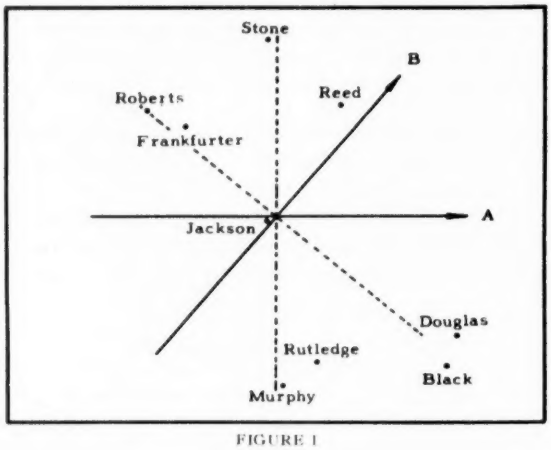

FIGURE 1

From such a model one could make some inferences regarding the similarities and dissimilarities among the judges. If two of the vectors were found to be fairly close together, such as 1 and 2, then one would infer that the two judges, Black and Douglas, have much in common in their respective points of view, as far as these can be inferred from the participation of these two judges in the dissenting votes of the Court. From rows 8 and 9 one can infer by inspection that the two judges, Frankfurter and Roberts, have much in common in their points of view as represented by the dissenting votes. Furthermore, since the vectors 1 and 2 are almost the collinear opposites of vectors 8 and 9, one would expect that Judges Black and Douglas have something in common in their points of view which is quite the opposite to whatever is in common to Judges Frank-

furter and Roberts. Other inferences of this nature can be made by inspection of the factor matrix in table 2.

The nine vectors in the factor matrix represent a three-dimensional configuration and it should be especially noted that the three coordinate axes, I, II and III, represent an arbitrary orthogonal reference frame. The location of this reference frame in the configuration is determined arbitrarily by the particular method of factoring that has been used to analyze the dimensionality of the given correlation matrix in table 1. It is an important step in a factor analysis to locate a reference frame in the configuration which shall be meaningful for interpretation of the common factor variance. The given correlation matrix of table 1, which shows the correlation coefficients, can be interpreted geometrically as showing the

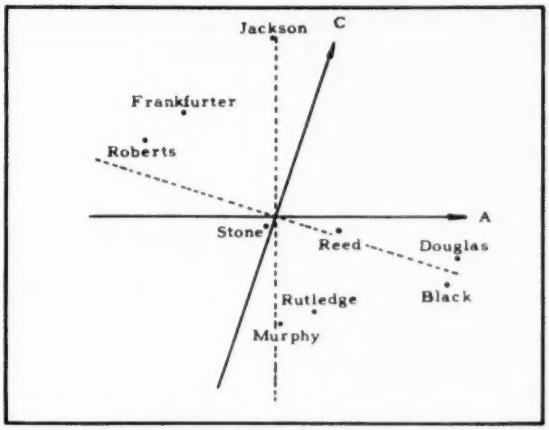

FIGURE 2

scalar products of pairs of vectors. It should be noted specifically that the scalar products of pairs of vectors do not define any reference frame.

It happens very frequently in factorial studies that the vectors which represent the variables arrange themselves in planes or in hyperplanes which are then chosen as coordinate planes. When such an arrangement is found in the configuration, we say that the structure is overdetermined and compelling as to the location of the reference axes and the coordinate planes. The combination of the vectors that represent the variables and the reference axes in such a case is called a simple structure. In the present case there are nine vectors in three dimensions and it is not to be expected that this small number of vectors will show a set of three clearly defined reference planes. An attempt has been made to use the same principles in the location of a preferred set of reference axes and these are represented

in the three diagrams of figures 1, 2 and 3. The authors do not insist that these reference axes are unique. Variant sets of reference axes could be found for this configuration. Table 4 shows the rotated factor matrix which represents the same configuration as tables 1 and 2. The transformation matrix of table 5 carries the structure from table 2 to table 4. The cosines of the angles in the reference axes of table 4 are shown in table 6. It will be noted that table 4 represents an oblique set of reference axes.

Figure 1 has been drawn to represent the relations between columns A and B of the oblique factor matrix in table 4. The factor A represents something which is shared by Justices Black and Douglas and which is opposite to what is common to Justices Frankfurter and Roberts. Factor B represents something that is common to the points of view of Justices Reed and Stone and which is opposite to what is common to Justices

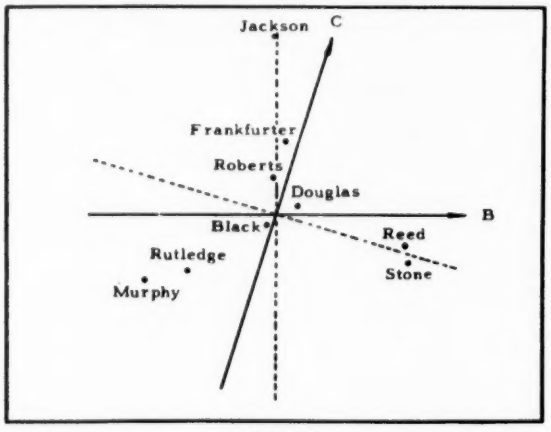

FIGURE 3

Murphy and Rutledge. Justice Jackson is indeterminate and unrepresented in the factors A and B, whatever they may represent. An alternative location of the reference frame in figure 1 is to use orthogonal reference axes if these are preferred. This would lead to a differentiation between two groups of judges. In one group there will be Justices Reed, Stone, Frankfurter and Roberts and on the opposite side would be Justices Black, Douglas, Rutledge and Murphy.

In figure 2 we have a graphical representation of the relations between factors A and C in the oblique matrix of table 4. Here we have another view of the same configuration. Again we see that factor A is strongly represented in Justices Black and Douglas and negatively represented in Frankfurter and Roberts. Factor C seems to be something that is represented by Jackson alone, although there is a strong component of the same

factor, whatever it may be, in Justice Frankfurter. Somewhat negative saturations of this factor are represented in the points of view of Justices Rutledge and Murphy. Justices Reed and Stone are not strongly represented in factors A and C, as seen in figure 2, whatever these two factors may represent.

In figure 3 we have a similar diagram for the relations between factors B and C. This view of the configuration shows again that Justice Jackson is conspicuous for factor C, whatever it may represent, and this factor is also represented in the point of view of Justice Frankfurter. The configuration in figure 3, in combination with the other two figures, suggests still another location of the reference axes for interpretation. The entire configuration of nine vectors might be thought of as representing actually five distinct attitudes or points of view which are linearly dependent, because the total configuration is only three-dimensional. The five groups would then be represented by the judges as follows, namely: (1) Reed

TABLE 3

THIRD FACTOR RESIDUALS

	1	2	3	4	5	6	7	8	9
1	(−0.04)								
2	−0.11	(−0.03)							
3	−0.05	−0.09	(0.00)						
4	−0.04	0.00	−0.01	(0.00)					
5	−0.07	−0.06	−0.03	−0.01	(−0.01)				
6	−0.12	−0.07	0.01	−0.05	−0.06	(−0.05)			
7	−0.02	−0.03	0.00	−0.01	0.00	0.02	(0.01)		
8	0.02	−0.02	−0.03	0.02	0.04	0.01	−0.02	(0.01)	
9	−0.08	−0.05	−0.12	−0.04	−0.11	−0.14	−0.04	0.01	(0.04)

and Stone, (2) Murphy and Rutledge, (3) Black and Douglas, (4) Frankfurter and Roberts and (5) Jackson and, to a lesser degree, Frankfurter.

The correlations between factors and the factor loadings in tables 2 and 4 are functions of the relative proportion of each type of case considered by the Court. For example, if one of the factors represented a division with respect to civil rights, and if only a few cases of this type appeared in the agenda of the Court, the effect on the correlations would be small and the factor would appear to be weak, although the actual division of opinion might be quite pronounced.

The dissenting opinions are not always in agreement. In a number of instances the majority opinion is accompanied by several dissents in which the announced dissenting reasons are different. The use of the intraclass coefficient puts all minority dissents in the same class. From the standpoint of social effectiveness, it is immaterial that rationalizations in the dissenting opinions differ, although we should remain cognizant of this circumstance in the interpretation.

It is the judgment of the authors that the interpretation of factorial studies in any field should be left to specialists in that field. In the present case one might proceed in one of two ways to arrive at some interpretation of the groupings of attitudes that are represented by the nine Supreme Court judges of the present study. Those who are familiar with the personalities of the judges and with the recent history of the Court might be able to agree, perhaps even spontaneously, about the interpretation of the factors that are involved here. The present factorial results have been

TABLE 4

OBLIQUE FACTOR MATRIX V

		A	B	C
Black	1	0.63	−0.03	−0.08
Douglas	2	0.67	0.08	0.05
Rutledge	3	0.14	−0.33	−0.31
Murphy	4	0.02	−0.49	−0.39
Reed	5	0.23	0.48	0.02
Jackson	6	−0.92	−0.01	0.64
Stone	7	−0.04	0.49	−0.04
Frankfurter	8	−0.35	0.04	0.27
Roberts	9	−0.49	−0.01	0.12

shown to some of our colleagues in political science, but in the discussions to date there have not appeared any interpretations that seemed to be unique and generally acceptable to all discussants. The results have also been shown to some of our colleagues in the law faculty with similar results, although there have been a number of individual judgments about the interpretation of particular factors. During these discussions we have made what seem to us to be rather interesting observations. We had expected to find that the factors would represent attitudes toward some general issues

TABLE 5

TRANSFORMATION MATRIX A

	A	B	C
I	0.42	−0.35	−0.34
II	0.77	0.92	0.00
III	0.48	0.18	0.94

TABLE 6

REFERENCE VECTOR COSINES C

	A	B	C
A	1.00		
B	0.65	1.00	
C	0.31	0.29	1.00

that might be called, for example, economic liberalism, or the rights of the individual, and such categories. When the results were discussed by some of our legal friends we had expected to find the factors interpreted in terms of some legalistic points of view. On the contrary, we found the lawyers discussing the factors in terms of the personalities of the judges. As psychologists, we had refrained from even considering the possibility that factors of personality and temperament would enter into any discussion by lawyers on this problem.

Perhaps a safer way to proceed in the interpretation of factors in this type of problem is to review carefully the 115 cases to make particular note of the judges who dissented in those cases and to classify them according to the agreements and disagreements of the particular judges. (A list of the cases can be supplied by the authors.) By examining the cases with grouping according to the votes of the judges it should be possible to clarify more objectively just what the factors are that are common to the cases on which the particular pairs of judges agreed and disagreed. Again, this is a study which should be carried out by lawyers and political scientists, rather than by the present authors.

In a previous study one of the authors[1] attempted to identify the blocks in a legislative body by the voting records of the members. The multiple factor methods have been developed since that time, and we are presenting here an illustrative example of the obverse factor methods in which correlations are determined between pairs of persons. The method seems to be promising for analyzing loyalties and identifications as identified by the voting records of the members of a group.

[1] Thurstone, L. L., "Isolation of Blocs in a Legislative Body by the Voting Records of Its Members," *J. Soc. Psychol.*, **3**, 425–433 (1932).

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